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#### PROGRESS REPORT

of the

## MARKET QUALITY RESEARCH DIVISION

#### AGRICULTURAL RESEARCH SERVICE

This progress report includes a summary of the current research of the Division and a preliminary report of progress made during the preceding year. It is primarily a tool for use of scientists and administrators in program coordination, development, and evaluation.

The summaries of progress on USDA and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed, will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations.

This report also includes a list of publications reporting results of USDA and cooperative research issued between July 1, 1968, and June 30, 1969. Current agricultural research findings are also published in the monthly USDA publication Agricultural Research. This progress report was compiled in the Market Quality Research Division, Agricultural Research Service, United States Department of Agriculture, Hyattsville, Maryland.

United States Department of Agriculture

Hyattsville, Maryland 20782

October 1, 1969



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#### INTRODUCTION

The research of Market Quality Research Division is concerened with the measurement, improvement, and protection of the quality of agricultural commodities in marketing channels. The work encompasses physiological, biochemical, pathological, and entomological problems encountered during the storage, transport, and distribution of agricultural commodities, and the development of new methods and devices for evaluation of quality.

Market Quality Research Division is a part of the Agricultural Research Service. It is headquartered at Hyattsville, Maryland. The greatest concentration of its scientific personnel is at Beltsville, Maryland, which includes the Instrumentation Research Laboratory, Color Research Laboratory, Postharvest Physiology Pioneering Research Laboratory, and personnel of Field Crops and Animal Products Research Branch and Horticultural Crops Research Branch. There are 28 field stations located throughout the country; 12 are located at state universities or branch experiment stations and two are in terminal markets.

Although a large variety of excellent quality fresh and processed agricultural products are retailed at reasonable prices throughout the year, there is need for further research on methods to reduce spoilage and waste during storage, transportation, and distribution, and to improve methods to reduce spoilage and waste during storage, transportation, and distribution, and to improve methods for evaluation of quality. Stored product insects and market diseases still destroy large amounts of produce regardless of costly controls. There is urgent need for new methods of control that will not create health hazards due to pesticide residues. Automated objective methods of quality evaluation are increasingly needed to make possible rapid reliable grading and inspection of large quantities of produce under modern packing, handling, and transporting conditions.

An appreciable amount of the Division's research is related to the effective performance of the service divisions of the Consumer and Marketing Service and the regulatory and control divisions of the Agricultural Research Service responsible for standardization, inspection, and grading of agricultural commodities. Market Quality Research Division also works closely with industry and other Government agencies on various problems relating to agricultural commodities in the marketing channels. Specific examples of recent research accomplishments by the Market Quality Research Division are:

## Sampling Shelled Peanuts

An automatic sampler has been developed that will take a sample from a moving stream of shelled peanuts prior to bagging. At present, samples of shelled peanuts are taken from bags with a slotted probe called a "trier." This method of sampling is not considered to be representative for grade purpose or for aflatoxin analysis. In addition the probing of individual bags causes considerable inconvenience to the sheller since each shipment of peanuts must be held so that all bags are accessible until the inspector has drawn a sample. Proper representative sampling of peanut shipments must be made to insure that the buyer and seller both receive correct payment for the peanuts sold.

## Meat Flavor

USDA scientists have developed an analyzer which is capable of resolving and quantitating accurately and speedily the amino acids present in beef muscle. The presence of and relative concentrations of amino acids or groups of amino acids present in beef carcasses during the postmortem aging contributes to or enhances the flavor of beef. It is estimated that about 5% of consumer acceptance of meat is based on flavor and aroma. Flavor grading of beef could yield about \$4.4 million annually to the producer.

### Trading Value of Vegetable Oils

USDA scientists have developed a more accurate method of determining total neutral oil of natural fats and oils. The total neutral oil value is important in trade channels because it determines the quality of vegetable oils and their market value in dollars. Several improvements resulted from the new method. It was more accurate, eliminated errors because it was semiautomatic, reduced costs and made possible the use of a non-air-conditioned laboratory.

## Automated Grain Grading

Experimental trials have demonstrated that a new automated test weight device is more rapid (15 seconds against 60 seconds), takes considerably less work and has comparable or better degree of precision than the manual procedure. Test weight is an important quality factor in grading grains. There is a constant demand for faster, more accurate measurements of grain quality and the development of an automated test weight device is a step in this direction.

#### Wholesomeness of Meat Products

Suitable tests are now available to detect horsemeat or meat of other unacceptable species in processed meat products. The tests have the advantage of little inter-species cross reactivity and can be used with heated products (frankfurters and bologna). The tests will aid the regulatory agencies in safeguarding the health of consumers.

### Shelf-Life of Milk

Research on pasteurized milk shows that bacteria apparently killed by heat can sometimes recover and grow normally under favorable conditions. USDA scientists are unlocking the mechanism of heat injury and the nature of the recovery at refrigeration temperature. This knowledge will lead to increased shelf life of pasteurized milk by our ability to inhibit the recovery mechanism.

### New Produce Storage and Transport Handbooks Issued

Up to date revisions of Agriculture Handbooks on "Protection of Rail Shipments of Fruits and Vegetables" and "The Commercial Storage of Fruits, Vegetables, and Florist and Nursery Stocks" were issued. These handbooks summarize many years of research for growers, shippers, and warehousemen and explain the proper temperature and handling conditions to prevent deterioration. Although many improvements in handling, storage, and shipping have been made, marketing losses for fruits and vegetables are still estimated to be at least \$220 million annually. Widespread use of the recommendations in the handbooks might conservatively result in a 10% (\$22 million) reduction of losses. The farm value of our fruits and vegetables is over \$3.7 billion.

## Hot Water Fungicide Treatments Reduce Decay

A combination of hot water and fungicide (Botran, 2,6-dichloro-4-nitroaniline) treatments give better control of postharvest decays of peaches, nectarines, and plums than either treatment used alone. The combined treatments (water at  $125^{\circ}$  + Botran at  $\frac{1}{4}$  1b/100 gallon for  $1\frac{1}{2}$  minutes) reduced decay of peaches and nectarines from 65% in untreated lots to about 8% in the treated lots. Plums required higher Botran concentrations ( $\frac{1}{2}$  1b/100 gallon), which reduced decay from 15% in untreated lots to 4% in the treated lots.

In 1968, 250 thousand tons of freestone peaches with a value of \$24 million were produced in California; 64 thousand tons of nectarines with a value of \$9.4 million and 108 thousand tons of plums worth \$18.7 million also were produced. Surveys in the New York and Chicago markets have shown that market losses in peaches, caused mostly by decay, amount to 15 to 24% through the consumer level. Losses in nectarines are probably a little higher than peaches and losses in plums a little less. Assuming an average loss of 20%, market losses in these three crops would amount to about \$10.4 million.

### Modified Atmosphere Doubles Storage Life of Avocados

Avocados stored in air keep satisfactorily for only about 1 month. By using storage atmospheres containing 1%  $\rm O_2$  and 9%  $\rm CO_2$  at 50° F, avocados can be stored satisfactorily for 2 months.

It is estimated that an increase of 1 month to the relatively short marketing season would increase the value of the avocado crop by \$1,800,000. The avocado industries will benefit directly and the consumer will benefit through a more steady supply of fruit.

## Forced Air Precooling Improves Market Quality of Strawberries

Strawberries can be precooled from high field temperatures to  $35^{\circ}$  F in  $1\frac{1}{2}$  to 2 hours by forced air cooling. Earlier procedures frequently required overnight cooling.

The faster cooling reduces decay, slows ripening, and makes it possible to deliver high quality berries to distant markets. This method of cooling also should help in expanding export sales.

There has been a dramatic switch to forced air cooling in 1968 and 1969. Several new facilities have been placed in operation and an estimated 90% of the strawberries shipped from California are now cooled by this method.

California produced 290 million pounds of strawberries in 1968, having a value of about \$61 million. Over  $\frac{1}{2}$  of the crop was marketed out of state as fresh fruit. Losses from decay and physical damage have been estimated at 20% through the consumer level, which would constitute a monetary loss of about \$7.6 million. Improved precooling along with refrigeration in transit would reduce the losses and result in delivery of a better product to the consumer.

USDA and experiment station research has contributed greatly to the acceptance and wide use of this new precooling method.

## New Wool Fabric Protectants Developed

Materials have been found which have a low mammalian toxicity, provide protection against moths and carpet beetles over a long period of time and continue in effectiveness even after the treated cloth has been washed or drycleaned one or more times. Among these are certain quaternary ammonium compounds and the insecticide Gardona.

Insofar as the general public is concerned, more individuals seek help in the control of fabric pests than for any other groups of insects. Damage to clothing, bedding, rugs, etc., is estimated to be in excess of \$350,000,000 per year. Everyone shares in this loss, although mills, marketing channels, and the military have a special concern.

The materials and methods developed have special interest in that they can be easily applied by the manufacturer, cleaning establishment, or by the individual in the home. They may offer protection for up to a year in storage and remain effective after laundering or drycleaning several times. They offer an alternative to the chlorinated hydrocarbon insecticides, the use of which may be restricted and for people who dislike the odor of moth crystals.

Adoption of these safe, long lasting, easy to use materials may boost the dropping per capita consumption of wool, since susceptibility to insects and the chore of protection is a distinct drawback in the public eye.

## Means Found to Protect Almonds in Storage

A method has been developed to protect almonds against insect infestation for the 9 to 12 months between harvest and utilization. It involves treatment at the time of storage with a quantity of malathion which will give the desired protection and leave no harmful residue.

The nation's almond crop, the annual value of which varies between 40 and 55 million dollars, is commonly infested with insects at the time it is placed in storage. This results from preharvest attacks and the crop is subject to continuing attack during storage. If no action is taken, about 50% of the almonds will become damaged or infested during the storage period. Direct losses through consumption by the insects and discard of infested or damaged nuts is estimated at between 1 and 2 million dollars per year. This would be higher if sorting and cleaning procedures costing additional millions of dollars annually were not used.

To treat the entire crop with malathion would cost less than \$5,000 per year.

## Method Developed for Killing Pink Bollworm in Cottonseed with Phosphine

The pink bollworm is regarded as the worst pest of cotton on a world-wide basis. Damage by this insect in the United States has been limited by vigorous measures to suppress infestation and prevent its spread. Larvae develop mostly in the seed and their spread, carryover, and reinfestation are associated with the ability of larvae to survive in the seed.

Larvae are destroyed when seed is processed, but will continue to survive in carryover unprocessed seed or seed held for planting. Fumigation of these is an essential part of the suppression and prevention program. Methyl bromide has been an effective fumigant but growers complained of detrimental effects on germination.

An alternative method, the use of phosphine, was developed. This involves the use of aluminum phosphide tablets which react with moisture in the air to produce the phosphine gas. All pink bollworms are killed by 120 hours of exposure to the gas produced from 60 tablets per 1,000 cubic feet under a polyethylene tarpaulin 1.5 mil or more in thickness in bulk or bagged seed. The fumigant effectively penetrates all types of bags used except unperforated polyethylene or saran bags 4 mil or more in thickness.

This method is of immediate use for treatment of all seed cotton for planting in the states of Arizona, New Mexico, Texas, Oklahoma, Nevada, and parts of Louisiana and Florida. It is also used for part of the bulk seed remaining at mills or gins during the "host free" periods.

Since the pink bollworm control program is a suppressive one, benefits are largely indirect. Losses in Hawaii when the insect was first found there when effective control was unknown were virtually complete causing abandonment of production. This treatment contributes to protection against a potential loss of a quarter of a billion dollars considering value of raw commodity alone. The magnitude of loss in textile and oil products and byproducts is many times greater.

## <u>Discovery of Cross Attractiveness of Sex Pheromones Opens New Avenues</u> for Control of the Khapra Beetle and its Relatives

In studies of sex attractant substances of several species of <u>Trogoderma</u> beetles, it has been determined that males of at least 2 species of pests of dried milk and grain are attracted by the khapra beetle female sex pheromone and male khapra beetles are attracted by the sex pheromones of the other 2 species. Work with additional species has indicated further interspecific attraction. It thus appears that one attractant, consisting of one or more chemical components, can be used to control several species.

The pheromones can be synthesized and offer possibilities for use as baits in traps to detect the presence and abundance or to eliminate males from the population, to "saturate" the environment and prevent mating by confusion, and to attract males to areas treated with insecticides or sterilants.

The khapra beetle is the world's most destructive pest of stored products. It is not now present in the United States but an infestation in 1953 in the southwestern United States required an expenditure of \$11 million to eradicate. Damage to dried dairy products by the other species and the cost of control now exceeds \$3 million per year. They also cause estimated annual losses of several million dollars in grain and cereal products. The findings reported here will be useful for control of possible future invasions of the khapra beetle.

## PROGRESS REPORT OF THE

# FIELD CROPS & ANIMAL PRODUCTS RESEARCH BRANCH MARKET QUALITY RESEARCH DIVISION AGRICULTURAL RESEARCH SERVICE

#### AREA 2

## DAIRY PRODUCTS

Research Activity Code: 2-50-707-14-204

2-50-702-14-214 2-41-412-14-220 2-41-501-14-221

## USDA and Cooperative Program

	:	Scienti	st	Man-Y	ears F.Y	. 1969
Location of Intramural Work	:_	Resear	ch	Prob1	em Area :	•
	:_	412	:	501	702	Total
	:		:		:	:
Maryland (Beltsville)	:	1.0	:	0.5	: 0.5	2.0
	:		:		:	•
	:		:		:	
Total	:	1.0	:	0.5	: 0.5	2.0
	:		:_		:	

Intramural program is supplemented by P.L. 480 funds in one country representing 7,855 U.S. dollars equivalent.

#### Progress - USDA and Cooperative Program

## RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

- 1. Moisture Distribution in Butter. Deterioration of print butter in cold storage (0°C or -20°C) was found to begin on the surface and to diffuse inward. A study was begun on the use of packaging materials impermeable to air (aluminum foil--parchment laminate). Results were inconclusive and samples were lost because of equipment failure before the study could be completed. (FC-89)
- 2. <u>Heat Stability of Bacteria</u>. Results obtained show unequivocally that some bacterial cells that appear killed by heat may recover and grow normally after incubation for long periods on a favorable medium. The

type of medium in which the bacteria are grown and heated, and the recovery medium used affect heat resistance and recovery. To obtain useful data on heat resistance of bacteria to be applied to food processing, the bacteria should be grown, heated, and allowed to recover in the food under study. Extrapolation of heat resistance data from one medium to another can be misleading. A new mathematical treatment was developed for the kinetics of thermal death of bacteria. It was shown that death results from inactivation of some fraction of multiple critical sites. The fraction of sites which must be inactivated to cause death apparently varies depending on the recovery medium used following heating. (FC-88)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. Analysis of Feeds and Forages for Chlorinated Pesticide Residues. Thin-layer plates incorporating vegetable oil and silver nitrate were found to be more color stable than other types of plates in which silver nitrate was incorporated. Plates incorporating only silver nitrate are unstable and it has therefore not been feasible to prepare them commercially since they cannot be kept for any length of time. The improved stability may make manufacture of commercial prepared plates feasible. The uniformity of the plates may also make it possible to analyse spots quantitatively using a thin-layer scanner. (FC-86)
- 2. Electrophoretic Patterns of Soluble Proteins in Bacteria. The project was inactive during the reporting period. (FC-115)

## RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- 1. Indicator Organisms in Dairy and Food Products. Results from 200 samples of raw milk from city markets and rural collection centers showed that the quality of market milk sold in Udaipur city was very poor. Mean sample plate counts at rural collection centers were 14,700,000/ml and at city market collection centers, plate counts were 110,000,000/ml. Two hundred and twenty samples of khoa were examined bacteriologically. Quality was very poor and the incidence of potentially enterotoxigenic staphylococcus was very high representing a real danger to the public health. (FC-99 (A7-MQ-13))
- 2. <u>Inhibition of Salmonella and Other Bacteria by Essential Oils</u>. The antimicrobial properties of citrus peel oils and derivatives against selected food-borne bacteria, either of public health significance or related to spoilage organisms, have been determined. More than 100 different types of bacteria were tested. Generally, terpeneless essential oils were bactericidal, while the other essential oils were bacteriostatic.

The use of 1,000 p.p.m. orange, lemon or grapefruit oils in skim milk resulted in a product more palatable than plain skim milk and with a shelf-life of more than 52 days at  $4^{\circ}$ C. The bacteriostatic effect of citrus oils decreased with increasing fat content of the milk products (0.1% B.F. to 12.0% B.F.). (FC-102)

#### Publications - USDA and Cooperative Program

### RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

- Dabbah, Roger, Moats, W. A. and Mattick, J. F. 1968. Some factors involved in the recovery of psychrophilic bacteria following sublethal heat treatment. J. Dairy Science 51(6):922. (Abstract). (FC-88)
- Dabbah, Roger, Moats, W. A. and Mattick, J. F. 1969. Factors affecting resistance to heat and recovery of heat injured bacteria. J. Dairy Science 52(5):608-614. (FC-88)
- Moats, W. A. 1969. Kinetics of thermal death of bacteria. American Society for Microbiology. Bacteriological Proceedings, p. 9. (Abstract). (FC-88)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Dabbah, Roger, Moats, W. A, Tatini, S. R. and Olson, Jr., J. C. 1969. Evaluation of the resazurin reduction 1-hour test for grading milk intended for manufacturing purposes. J. Milk and Food Technol 32(2): 44-48. (MQ 3-44)
- Moats, W. A. 1969. Note on the use of vegetable oil as a source of peroxide in thin layer plates for analysis of chlorinated pesticides. J. Association of Official Analytical Chemists 52(4):871-872. (FC-86)

#### AREA 4

#### GRAIN

Research Activity Code: 1-38-702-14-201

1-38-501-14-209 1-38-408-14-210 1-38-501-14-215

#### USDA and Cooperative Program

	:Scientist Man-Years F.Y. 1969							
Location of Intramural Work	: Research Problem Area :							
	: 408	: 501	: 702	: Total				
	:	:	•	•				
Maryland (Beltsville)	: 0.5	: 2.4	: 0.3	: 3.2				
Kansas (Manhattan)	: 1.0	: 2.0	: 0.0	: 3.0				
Texas (College Station)	: 0.0	: 0.7	: 0.0	: 0.7				
	:	:	:	:				
	:	:	•	•				
Total	: 1.5	: 5.1	: 0.3	: 6.9				
	:	:	:	:				

Intramural program is supplemented by extramural support representing (a) 1.7 SMYs at State Agricultural Experiment Stations  $\frac{1}{2}$ , (b) 3.3 SMYs at other U. S. institutions  $\frac{2}{2}$ , and (c) P.L. 480 funds in three countries representing 56,447 U. S. dollars equivalent.

## Progress - USDA and Cooperative Program

## RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Control of Microorganisms in Stored Grain. Of 12 chemicals tested as mold inhibitors in grain, propionic acid was most effective. The high rates of application required would make commercial treatment expensive, probably equivalent to drying costs. Mold-produced enzyme products sold as silage and high moisture grain preservatives contained low microbial populations and did not have any apparent effect on grain in laboratory storage trials. Due to weather conditions, on-farm storage tests were not conducted. A quantitative beta-carotene test

<sup>1/</sup> RPA 501 1.2; and RPA 702 0.5

<sup>2/</sup> RPA 501 3.3

as an indicator of fungal invasion of corn was developed. Evaluation of the test is incomplete; it may be too time-consuming for practical use.

When the moisture content of grain sorghum is adjusted to 18-24%, subsequent fungal growth is the same whether inoculated with such fungi as Rhizopus, Mucor, Alternaria, and Fusarium. The moisture requirements of these fungi cannot be studied in autoclaved or chemically sterilized grain because they grow too easily on dead seeds with no competition from other fungi. Treated seed from which the fungicide has been washed was found to be a fairly satisfactory material for these studies. The seeds are alive, but apparently free of fungi. Presently seeds inoculated with various fungi alone, or in combination, at various moisture contents are being studied. (FC-124)

- 2. Changes in Quality Indicators as Corn and Wheat Are Stored Under Various Conditions. Equipment for storage tests was set up. Corn and wheat samples were obtained for preliminary tests. Progress was made in developing techniques for sample treatment under varying conditions of temperature and atmosphere (N2, O2, and combination). Corn and wheat of the 1969 crop will be used for tests. (FC-119)
- 3. Origin and Characterization of Mustiness in Grain. Nineteen strains of Aspergillus niger were used in this investigation. The strains differed in morphological and growth characteristics. Standard conditions were used in the studies to produce mold odor: Coarse wheat meal was incubated with the mold for 72-84 hours at 28-30°C and 92-96% relative humidity. The odors could be classified into six defined types; namely, fusty, fungal, fruity, mashy, herbal and soily. Gas chromatograph studies will be made of the other odor fractions and the chemical compounds will be identified if possible. (FC-96 (E21-MQ-2(k))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Automatic Test Weight Device. Test weights on 100 samples each of corn and wheat have been completed. Test weights on these 100 samples were made by two different operators, in duplicate, on both the manual and the mechanical device (800 analyses total). The time required to obtain the weight per bushel with the mechanical device is approximately one-half the time required by the manual method; also, less labor is involved. Data for the wheat samples showed a difference between the two operators and the two test weight devices. The standard deviation between duplicates for the manual and mechanical method were 0.08 and 0.10 lbs/bu., respectively. (FC-31(C))

- 2. Sound Grain Determination. Wheat samples damaged to varying degrees by soaking and drying showed a correlation between degree of damage and the number of internal cracks when examined by X-ray microradiograph. A direct relationship between the vapor pressure of water over heatdamaged samples and their percent moisture content occurred at temperatures above 42°C. The UV spectra of ethanol extracts of wheat samples impregnated with molds showed significant differences between sound and moldy samples. Identical spectra were observed for various sound grain extracts. The method is complicated by the adsorption at 275 mm by fungal metabolites which mask the absorption by the sound grain component. Differences occur in adsorption rates between different types of damaged corn and between different levels of a specified damage. Adsorption curves follow the Elovich adsorption kinetic equation. Relative humidity, initial moisture content, and temperature, all influence adsorption rates. Laser beam reflectance measurements along the circumference of four sample classes of wheat kernels were completed. Maximum reflectance values exhibit a greater spread than minimum values. (FC-32(C))
- 3. Optical Characteristics of Grains. Reflectance data for 45 samples each of twelve different grains were analyzed and the wavelengths determined at which it is theoretically feasible to discriminate admixture grains from the primary grain in certain grain samples. Little difference existed between reflectance curves of hard red spring and hard red winter wheats and between curves of barley and oats. It does not appear feasible to separate expected admixture grains from grain sorghum. Photographs were taken from which dimensions of individual kernels were obtained. These data will be analyzed to determine if significant differences exist in the profiles of different kernels and to determine the variation of the profile of kernels within the same grain. The profile data and the reflectance data will be analyzed in connection with each other to determine if this type of data can be used to identify certain grains. (FC-33(C))
- 4. <u>Isolating Germ of Grains</u>. A bleaching method developed shows considerable promise in reducing the time for observing germ damage in grains. Tests with several grain inspectors in Kansas show the time required by the bleaching method as compared to the standard method is approximately one-third. Removal of the hull from oats and barley with a Foresberg Impact Dehuller was unsuccessful. The Foresberg Sample Seed Scarifier will remove the hull from barley. (FC-34(C))

- 5. Automatic Moisture Meter. Individual grain kernels were run through rollers of a modified Tag-Heppenstall machine to evaluate resistance techniques. Voltage representing a function of grain resistance was recorded for each kernel and plotted against oven determined moisture. A correlation existed between the two determinations but in general the scatter in values was not superior to results obtained using a capacitance chamber method. In an attempt to reduce the influence of grain size, grain geometry, and contact pressure on moisture readings, a probe method was devised for placing small electrodes inside the kernel. The data was encouraging but because of technical problems it was concluded not to be a feasible method. Data show that surface moisture affects conductance-type measurements more than capacitance-type measurements. However, considering all data, electronics and mechanics of a device, it appears that the best system for a histogram-generating moisture instrument is a resistance type system utilizing rolls.
- 6. Objectionable Odors. A quantitative method for analyzing volatile compounds from grains was developed. With proper control of conditions excellent repetitive results of samples derived from duplicate aliquots of the same grain sample can be achieved. Approximately 70 corn samples were analyzed and results were subjected to preliminary statistical analysis with various computer programs. These preliminary analyses showed that 16 of the 70 samples should be removed from the analysis for various technical reasons. The final analysis was made on 54 sets of data consisting of 14 no odor, 17 sour and 23 musty corn samples. A computer discriminate analysis of the data properly classified 86 percent of the 54 samples. Of 7 samples misclassified, 3 were test cases and were expected to cause problems. Accumulation of odor data for wheat and grain sorghum evaluation is in process. (FC-103(C))
- 7. Detection of Internal Insects. Ten reagents for detecting amino acids, six reagents for detecting quinones and one reagent each for detecting trehalose and phenols, and twenty-two media were evaluated. A ninhydrin-collidine reagent for detecting amino acids appears to be the best reagent because it has a strong blue color, high sensitivity, and a low blank color. The reagents for detecting quinones, the reagent for detecting trehalose and the reagent for detecting phenols are not promising. Millipore phoroslide is promising as a medium because of its low blank values and durability. Fluorescence of insect body fluids is promising because of its sensitivity and reproducibility. (FC-104(C))

- 8. General Appearance. The purpose of this project is to develop an objective method of determining general appearance of grains. A Boerner sample divider was evaluated to assure that it showed no bias in dividing samples. An extensive literature review was started to provide resource information on alternative procedures and techniques. The Hunter color difference meter was installed and standardized. The circumferential lighting of the large optical unit was inaccurate in the redness range. This is being corrected. (FC-134(A))
- Kernel Hardness. High multiple correlation coefficients were obtained between various kernel hardness indices and most all of the technological properties tested. In most cases, these high R-values resulted from the addition of terms describing the effects of moisture content and kernel size or kernel weight in the multiple regression The contribution of kernel weight was greatest in the combination with the pearling or the temnometer test, and fairly great in the comprimeter test; the contribution of moisture content was largest in combination with the durograph test or the comprimeter test. The MIAG- and durograph-particle size indices were best kernel hardness tests for predicting technological properties; the pearling and the comprimeter tests may be of value. Some practical conclusions were: (1) Brabender Durograph and Strong-Scott barley pearler are most practical devices to determine kernel hardness. (Pearling test is subject to variation in kernel size.), (2) high degree of correlation between kernel density and percent vitreous wheat, (3) larger kernels yield most flour, use least power, and (4) generally, addition of moisture softens wheat, reduces kernel hardness (except barley pearler method). Added moisture also reduces flour yield, minimizes bran and ash contents, and generally yields smaller flour particles. Optimum differentiation between hard and soft wheat was obtained at approximately 10% moisture. (FC-21 (E19-MQ-1(a))
- 10. Determining Proportions of Durum and Common Wheat in Semolina Products. Four biochemical interspecific methods were found for detection of common wheat in durum wheat products: (1) Sitosteryl palmitate content (not practical), (2) electrophoretic components from water-soluble protein (tedious), (3) total petroleum ether protein fraction (tedious), and (4) an electrophoretic component from chloroform-methanol protein (tedious). The chloroform-methanol protein method was the only method with which there were no exceptions in detecting types of flour. In 84 varieties tested, the chloroform-methanol protein results in common wheat varied from 53-104; in durum,

it was barely detectable (0-21). In manufacturing pasta the petroleum ether protein method showed significant differences between varieties; whereas, the other three methods were not significantly affected by variety. A general survey of common and durum wheat varieties from different countries was made to set tentative specific limits of variations for the four methods. Based on these limits, the minimum and maximum content of common wheat in an unknown mixture is calculated as a function of each of the four methods. A comparison of the four methods, based on the intervals of uncertainty of the estimation, indicates that the best results are obtained by the method based on the chloroform -methanol protein method. These results can be improved by concurrent use of the four methods. (FC-22 (E25-AMS-7(a))

- 11. Hidden-Insect Detection in Grain. A machine, employing a vibratory feeder and vacuum pickup wheel, was designed, constructed, and tested for transporting individual kernels of grain over an X-ray scanning device. Tests of the X-ray scanning device showed a high degree of accuracy in the automatic determination of infested vs. non-infested kernels. These tests also showed that a highly stable X-ray beam stabilizer is necessary in order to make the automatic scanning operation practical. An X-ray anode current stabilizer employing a photoelectric-sensing device in conjunction with a silicon-controlled rectifier controller was designed and constructed. This device reduced X-ray energy drift from over 30% per hour to less than 3% per hour. (FC-129)
- 12. Evaluation of the Quality of Gamma-Irradiated Cereal Grains. Samples of nine lots of hard red winter wheat which had been infested with insects, stored, and then irradiated at about 27 and 41 krads were received for milling and baking analysis. Pre-treatment and post-treatment samples were received. Wheat samples have been milled, aged, and bread baked from the flour. Other quality tests are being processed. (FC-27)
- 13. Relation of Lipids to Grain Soundness and Quality. Compositional differences were studied in lipids extracted by petroleum ether from corn and wheat at moisture contents near 10, 12 and 16%. In wheat the percentage of lipid extracted increased as the moisture content was increased. This consistent increase was not observed in corn. The ratio of free fatty acids to total lipid content was higher in both corn and wheat at 16% than at 10% moisture content, and more polar lipids were extracted at the higher moisture content. When the moisture content was raised, percentages of nitrogen and phosphorus

indicated that more phospholipids and lipoproteins were extracted from wheat and more phospholipids were extracted from corn. Changes were noted in the mixing characteristics of wheat flour as related to the presence and absence of the lipid fraction. In general, the mixing stability of the flour is less when the lipid fraction is removed. Damaged lipids resulting from deteriorated germ in wheat was also noted to adversely affect the baking quality of the wheat. (FC-28)

- 14. Mechanical Sampling of Grains. High speed movie films of a diverter traversing a grain stream indicate that while the diverter discharges all the way across, it appears to discharge most heavily near the middle of its course of travel. Side views through a plexiglass diverter mounted in a stationary position under a spout show distinct differences in the velocity of the grain moving through the diverter. Side views of a plexiglass diverter attached to the transporter picturing the diverter as it moves out of the grain stream towards the cameras show that the diverter is quite full after cutting the grain stream. Films showing a side view of the diverter as it cut completely through the grain stream shows that a distinct layer of grain builds up in the bottom of the plexiglass diverter after traversing one-half of the grain stream. Preliminary tests of determining percentages of different grains in a mixture by neutron activation were made. (FC-36)
- 15. Grain Breakage Test. Further tests were made during the 1968 crop year using the Stein grain breakage tester. Two Stein units were compared using corn, soybeans, wheat and grain sorghum. Results showed that the Stein unit, with certain modifications, would be a suitable device for a standard of comparison in a grain breakage test. Recommendations have been made for the development of a standard grain breakage index. (FC-113)
- 16. Variation in Yeast and Its Effect on the Experimental Baking Test. Work was concerned with the development of a simple test to measure variation in different brands of commercial yeast. Since fermentation power of the yeast is its most important attribute the test developed measured CO<sub>2</sub> from a fermented standard medium. The following medium was found best: 4 g. sucrose, 2 g. dextrose, 1 g. salt, and distilled water to make 100 ml. solution. The test consists of shaking 6 g. of compressed yeast or 3 g. of active dry yeast in a flask with 25 ml. of distilled water; then 100 cc. of medium are added and the solution fermented in a controlled temperature bath at 86°F. for 4 hours.

The experimental error in the test is slight; duplicate results check well. A substantial variation was found among different brands of compressed and active dry yeast. Such difference might cause problems within bakeries if the brand of yeast were changed without corresponding changes in the baking formula. (FC-116)

- 17. Sample Divider. The prototype electric programmed control circuit for this divider has been constructed. Preliminary testing of the circuit indicates it is satisfactory. Construction of the mechanical portion of the weighing system is progressing. This device will be used to automatically divide grain samples into a pre-set number of subsamples of predetermined weights. This will eliminate manual dividing and weighing of samples in a laboratory for different individual analyses. (FC-117)
- 18. Accurate and Rapid Objective Tests for Evaluating Quality of Bread Crumb Grain and Crumb Color. No work has been done on this project during this reporting period. (FC-125)
- 19. Estimating Weevil Damage in Peas. Peas containing weevil larvae have a lower density than uninfested peas. A method has been developed for estimating extent of weevil damage in peas. The method is based on measurements of the percent of peas that will float on a sodium chloride solution having a certain specific gravity. (FC-138)

## RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

1. Fungal Contamination and Metabolites in Grain. A nondestructive technique for detecting mold contamination in single kernels of corn by fluorometry was developed. This technique is based on the fact that the fluorescence from the kernel is quenched or reduced on mold-contaminated corn. Four optical property measurements were compared to determine which technique would be most effective in distinguishing between sound and mold-contaminated corn. The best criterion of mold contamination in corn kernels was the ratio between two measurements of the fluoresced energy (at 442 nm and 607 nm). The results showed that the contaminated kernels could be distinguished from the sound kernels with less than 5% error. Our rapid technique for screening corn for aflatoxin by assaying corn dockage, reported previously, was evaluated. Aflatoxin was found in every dockage portion tested. Corn dockage samples contained an average of about six times more aflatoxin than dockage-free corn samples. (FC-18)

- 2. Mycotoxin and Dryeration Studies on Corn. Corn harvested at 30% moisture was dried to 20% with heated air and 18% by aeration. High moisture harvesting resulted in large amount of mechanical damage and fine material. The grain molded in storage, even when aerated with 0.5 cfm/bu. Harvesting at 25% moisture and dryerating to 16-17% moisture resulted in little mold damage during storage with airflows of 0.3-0.5 cfm/bu. The corn stored well when dryerated to 18% moisture and then aerated with 0.7 cfm/bu. Isolates of Aspergillus flavus and A. parasiticus required minimum relative humidities of 81-82% for germination, 82-83% for sporulation, and 84-85% for aflatoxin production on corn. Precise minimum corn moisture contents were not established for A. flavus activity. Isolates of Penicillium viridicatum were toxic to test animals resulting in: Reduction in weight gains, depression, high mortality and pathologic alternations of liver and kidneys. High doses of tetracycline antibiotic added to toxic diets reduced severity of symptoms; same high doses were harmful when added to non-toxic diet. Heat treatment at 100°C or higher for 10 minutes reduced but did not eliminate toxicity of fungal cultures. is present in mycelia but not culture filtrates from liquid cultures: It is extractable in hot chloroform. (FC-82(A))
- 3. Relation of Microflora to Quality of Grains and Seeds and Their Products. Temperatures of 10-12°C were very effective in preserving quality of Grade No. 2 corn with a moisture content of 14.5-15.5%. Deterioration was rapid at 25°C. Rice, with 16% moisture, germinated over 90% after storage for 465 days at 5°C. Germinability and invasion by storage fungi were much slower at 15° than at 25 and 30°. Deterioration of rice, even in lots of relatively high storage risk, was greatly reduced by storage at 5-15°. Incidence of Aspergillus flavus did not increase in rough rice with moisture contents below 18%, even at moisture levels and temperatures that should favor an increase. Apparently A. flavus could not compete with other species of Aspergillus. These studies showed that moisture content of sunflower seeds of 10% for Grade No. 1 and 14% for Grades 2 and 3 are too high, especially for seeds of high oil content, such as the variety Peredovic. A temperature of 10°C greatly reduced invasion of sunflower seeds by storage fungi. A rating system was developed to evaluate storability of rice based on germinability and numbers of colonies of storage fungi per gram of grain. Mycotoxin studies were conducted on the conditions necessary for the production of the estrogenic compound, F-2, by various isolates of Fusarium roseum. (FC-17(G))

- 4. <u>Investigations on the Aflatoxin Analysis in Grain Crops</u>. Grant became effective in February 1969; therefore, no progress report has been received. (FC-131 (E21-MQ-3)
- Refrigerated High Moisture Corn. Field shelled corn with 20% 5. and 25% moisture content and an initial temperature of about 85°F was placed in bins with no aeration during the first 12-16 hours. The corn was then cooled to 35-40°F in 2, 4, or 8 days by ventilating at 0.5 cfm/bu with air of various temperatures. The overnight delay in cooling permitted rapid initiation of mold growth and a rise in temperature. At 25% moisture the 2-day cooling rate resulted in little mold growth and no apparent changes in fat acidity value (FAV) or germination percentage. The 4-day cooling rate permitted some deterioration in the middle and top of the bin, and the 8-day cooling rate resulted in deterioration (mold growth, increase in FAV, decrease in germination) throughout the bin. Aspergillus flavus was the principal storage mold involved, and aflatoxin in concentrations up to 95 ppb was found in the slow cooled bin and in the upper part of the bin cooled in 4 days. At-20% moisture only the 8-day cooling rate resulted in any appreciable mold growth, with Aspergillus glaucus dominating. No losses in germination were detected, and FAV increased slightly in the top of the slow cooled bin only. Holding the corn at 70, 85 or 100°F from harvest up to 4 days resulted in appreciable mold growth and deterioration only after 2 or more days at 85 or 100°F. The corn held at 25% moisture and 85°F for 4 days contained 200 ppb of aflatoxin  $B_1$ . (TF-2-057B)

## Publications - USDA and Cooperative Program

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

Garcia-Faure, R., Garcia-Olmedo, F. and Vallejo-Acevedo, J. M. 1968.
Estimation of <u>Triticum aestivum</u> in pasta flours: Interspecific limits for sitosteryl palmitate content. J. Sci. Fd. Agric. 19:322-324. (FC-22 (E25-AMS-7(a))

Greenaway, W. T. 1969. A wheat hardness index. Cereal Science Today 14(2):4-7. (Related to FC-21 (E19-MQ-1(a))

Kramer, H. A. 1968. Performance of three diverter-type mechanical grain samplers and the pelican. USDA, ARS 51-16. (FC-36)

Kramer, H. A. 1968. Sampling of wheat, soybeans, and corn transported in covered hopper cars. USDA, ARS 51-20. (FC-36)

Kramer, H. A. 1968. Effect of grain velocity and flow rate upon the performance of a diverter-type sampler. USDA, ARS 51-25. (FC-36)

## RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- Birth, Gerald S. and Johnson, Robert M. 1968. Detecting mold contamination incorn by optical measurements. Presented at Amer. Soc. Agr. Eng. Winter Meeting, Chicago, Ill. Paper No. 68-566. (FC-18)
- Johnson, Robert M., Greenaway, Walter T., and Golumbic, Calvin. 1969. Sampling stored corn for aflatoxin assay. Cereal Sci. Today 14(2): 25-29. (FC-18)
- Sauer, D. B. and C. M. Christensen. 1968. Germination percentage, storage fungi isolated from, and fat acidity values of export corn. Phytopathology 58:1356-1359.(FC-17(G))
- Sauer, D. B. and C. M. Christensen. 1969. Some factors affecting increase in fat acidity values in corn. Phytopathology 59:108-110. (FC-17(G))

AREA 4a

RICE

Research Activity Code: 1-38-702-14-201

1-38-501-14-209 1-38-408-14-210 1-38-501-14-215

### USDA and Cooperative Program

	:Scientist Man-Years F.Y. 1969							
Location of Intramural Work	:_R	esear						
	:	501	:	702	:	Total		
	:		:		:			
Texas	:	1.1	:	2.0	:	3.1		
	:		<u>:</u>		:			
	:		:		:			
Total	:	1.1	:	2.0	:	3.1		
	:		_:		<u>:</u>			

Intramural program is supplemented by extramural support representing (a) 0.5 SMY at State Agricultural Experiment Stations  $\frac{1}{4}$ , and (b) P.L. 480 funds in three countries representing 33,675 U. S. dollars equivalent.

## <u>1</u>/ RPA 501 0.5

## Progress - USDA and Cooperative Program

## RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Storage Changes in Milled Rice. The influence of protein content and distribution on the cell contents and properties of cooked rice were studied using untreated and trypsin treated rice. Microscopical study of structure showed that kernel integrity was maintained after cooking when a high protein content was found in the outer layers of the kernel. Initial results of studies in progress verify that proteins are chemically modified by reactions with breakdown products of lipid oxidation; such modifications are associated with quality changes. No parallel change in the starch fraction was observed.

Samples of 21 rice varieties have been milled and graded. Aliquots were stored at 25°C and will be studied after an aging period of 3 to 4 months. Aliquots of all samples were characterized before storage as a basis for measuring changes in quality and the effect of such characteristics on quality changes. Significant differences in alphamino N were found between rice varieties and their proteins. The alpha-amino N liberated by typsin hydrolysis is one of the parameters of the proteins showing changes during storage of rice. (FC-73 (E25-AMS-9(a))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Degree of Milling and Color of Rice. Cooperative tests with the Grain Division, C&MS, of the Rice Ratiospect instruments have shown that two or more instruments are difficult to keep in calibration. It has not been determined if this is due to differences in optical and photoelectric components or due to changes in the electronic components or circuitry. Tests of an Icore small particle color sorter show that it is efficient in separating red rice and dark heat-damaged rice from normal rice but rather inefficient in separating light, heat-damaged, chalky and nontranslucent rice from normal rice. Our studies have shown that transmitted light is more efficient for separating opaque rice kernels from normal, translucent kernels than reflected light. (FC-91)

## RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

1. Microbiological, Chemical, and Physical Deterioration of Rough Rice. In rough rice stored in a relative humidity of 90% or greater, Aspergillus candidus invaded and competed more successfully than A. parasiticus. The percentage of seed infected by A. parasiticus was reduced as much as 90% by inoculating rough rice with A. candidus and A. parasiticus spores at ratios approaching 1:1. Species of the A. glaucus group became the most prevalent fungi infecting stored rough rice at equilibrium with relative humidities of 90% or less. Differences in the ability to infect rough rice stored at different levels of relative humidity were found between strains of Aspergillus repens. A xerophytic strain infected 10 to 20 times as many seeds at low relative humidities as the other strains tested. A significantly higher level of infection by this strain was also maintained during extended storage. Infection by storage fungi was greatly reduced in

stored high-moisture tough rice by controlling temperature with refrigerated air compared to air at ambient temperatures; deterioration as measured by loss of grade proceeded at about the same rate under both conditions. (FC-75)

2. Mycotoxins in Rice. Samples used in this study consisted of milled rice from representative areas of Japan sampled in 1966. The toxicity of 144 representative strains, including 50 species of Penicillium and 45 species of Aspergillus were tested for toxicity by means of forcefeeding to mice of culture filtrates. In this test, five species, P. viridicatum, P. expansum, P. simplicissimum, A. repens, A. versicolor, and Cladosporium cladosporioides caused the death of all test animals. In total, 53 (ca 37%) of the strains were toxic or mildly toxic. The latter was indicated by stunting of growth or weight loss. Damage to the brain, liver, and kidneys was associated with toxicity by histopathological studies. Of 22 genera in the test only 8 revealed no toxic strains. Furthermore, only 1 to 3 strains of many genera were included in the test. The tests also demonstrated a significant strain-substrate interaction. Some strains produced toxic metabolites on one substrate but failed to do so on another. P. roqueforti did not produce toxic metabolites. Three toxic fractions were isolated from culture filtrates of P. roqueforti. (FC-74 (A11-MQ-2))

From the 1968 crop, 127 cultures of the Aspergillus flavus group have been tested for production of aflatoxin on a rice substrate. A preponderance of non-aflatoxin-producing strains of the  $\underline{A}$ . flavus group was found. The prevalence of  $\underline{A}$ . oryzae and many isolates with characteristics between  $\underline{A}$ . oryzae and  $\underline{A}$ . flavus increased in the 1968 rice crop. In a pilot-scale study of rough rice stored at moisture contents up to 16.9% (wet basis) under aeration for up to 184 days, aflatoxins were detected in only one of 62 samples. Strains of  $\underline{A}$ . flavus spp. were isolated from a significant percentage of the kernels of all samples throughout the duration of the storage test. (FC-78)

The internal and surface-borne microflora of rice have been isolated and identified. About 632 samples of grain, including rice, have been examined for microflora and their degradation products. Metabolites of <u>Aspergillus candidus</u>, a species often infecting rice, have been found to be toxic to birds and rats. The analysis of the metabolites of other species of fungi infecting rice is in progress. The

relationship between stored-products insects and storage fungi have been studied. Insects were shown to be carriers of fungal inoculum. Experiments designed to clarify the role of insects in fungal deterioration of rice in large storage structures are in progress. Extensive studies on the action of the volatile chemicals and fumigants and fungicidal effects were conducted. Organic acids were not effective in inhibiting internally-borne fungi below 1%. Synergism of ammonia and phosphine on the fungicidal effect was established. (FC-98 (A7-MQ-12))

3. <u>Development of a Rice-Sizing Device</u>. Louisiana State University is conducting research to develop a device, which will, in one operation, automatically sort and determine the percentages of each length classification of milled rice. Research to date has resulted in the development of an optical device capable of discriminating between particles differing in length by as little as .005". Attempts to develop a high-speed handling and orienting device to complement the optical device have been unsuccessful thus far. (FC-123(C))

## Publications - USDA and Cooperative Program

## RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

- Barber, S., C. Benedito de Barber and J. L. Guardiola. 1968.
  Almacenamiento de arroz elaborado. V. Cambios en los azucares segun la localiz ación en el grano. Agroquimica y Tecnologia de Alimentos. 8:89-101. (FC-73 (E25-AMS-9(a))
- Hernandez, E., R. Vila and J. Garcia de la Cuadia. 1968. Microflora del arroz elaborado. 1. Flora de molinos industriales. Agroquimica y Technologia de Alimentos. 8:240-248. (FC-73 (E25-AMS-9(a))
- Primo, E., S. Barber, C. Benedito de Barber and L. Sanchez. 1968.
  Almacenamiento de arroz elaborado. IV. Modificación de los
  componentes nitrogenados. Agroquimica y Tecnologia de Alimentos.
  8:224-230. (FC-73 (E25-AMS-9(a))
- Primo, E., S. Barber, C. Benedito de Barber and L. Sanchez. 1968.
  Almacenamiento de arroz elaborado. VI. Cambios de los
  componentes nitrogenados segun la localización en el grano.
  Agroquimica y Tecnologia de Alimentos 8:231-239. (FC-73 (E25-AMS-9(a))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Stermer, R. A. and Schroeder, H. W. 1968. Market quality research of rice, 1967-1968. Rice Journal 71(7):68-69. (FC-91)
  - RPA 702 PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
    ANDNNATURALLY OCCURRING TOXINS
- Boller, R. A. and H. W. Schroeder. 1968. Accumulation of aflatoxins in stored rice in relation to competition between <u>Aspergillus</u> parasiticus Speare and <u>Aspergillus chevalieri</u> (Mangin). Thom and Church. Proceed. Assoc. of Southern Agri. Workers. 65th Annual Convention. p. 204 (abstract). (FC-78)
- Kurata, H., S. Udagawa, M. Ichinoe, Y. Kawasaki, M. Tazawa, J. Tanaka, M. Takada and H. Tanabe. 1968. Studies on the population of toxigenic fungi in foodstuffs. V. Acute toxicity test for representative species of fungal isolates from milled rice harvested in 1965. J. Food Hygienic Soc. (Japan) 9:379-384. (FC-74 (A11-MQ-2))
- Kurata, H., S. Udagawa, M. Ichinoe, Y. Kawasaki, M. Tazawa, H. Tanabe and M. Okudaira. 1968. Studies on the population of toxigenic fungi in foodstuffs. VI. Histopalthologic changes in mice caused by toxic metabolites of fungi from domestic rice. J. Food Hygienic Soc. (Japan) 9:385-394. (FC-74 (A11-MQ-2))
- Raghunathan, A. N. and S. K. Majumder. 1967. Problems of fungal disorders in stored grain. Symposium on diseases during storage and transit and control. J. Ind. Phytopathological Society, Varanasi, 1967. (FC-98 (A7-MQ-12))
- Schrodder, H. W. 1968. Aflatoxin development in undried rough rice. Proceedings of the 1967 Mycotoxin Research Seminar. p. 93-95. Washington, D. C., June 8-9, 1967. Issued Sept. 1968. USDA. (FC-78)
- Schroeder, H. W., R. A. Boller, and H. Hein, Jr. 1968. Reduction of aflatoxin contamination of rice by milling procedures. Cereal Chem. 45:574-580. (FC-78)
- Schroeder, H. W. and Hugo Hein, Jr. 1968. Effects of diurnal temperature cycles on the production of aflatoxin. Appl. Microbiol. 16:988-990. (FC-78)
- Sugujama, J., Y. Kawasaki, and H. Kurata. 1968. <u>Wardomyces simplex</u>, a new Hyphomycete from milled rice. The Botanical Magazine (Tokyo) 81:243-250. (FC-74 (A11-MQ-2))

#### AREA 4b

#### FEED AND SEED

Research Activity Code: 1-38-501-14-205

1-38-408-14-206 1-38-501-14-215

## USDA and Cooperative Program

	:Scientist Man-Years F.Y. 1969 :_Research Problem Area_:						
Location of Intramural Work							
	:	408	:	501	:	Total	
	:		:		:		
Maryland (Beltsville)	:	1.6	:	4.9	:	6.5	
	:		:		:		
Texas	:	0.0	:	0.2	:	0.2	
	:		:		<u>:</u>		
	:		:		:		
Total	:	1.6	:	5.1	:	6.7	
	:		:		:		

Intramural program is supplemented by extramural support representing (a) 0.3 SMY at State Agricultural Experiment Stations  $\frac{1}{4}$  and (b) P.L. 480 funds in four countries representing 67,000 U. S. dollars equivalent.

## 1/ RPA 408 0.3

## Progress - USDA and Cooperative Program

#### RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Relation of Fungi to Quality of Seeds, Grains and Their Products. In a study of the effect of seed vigor level upon time of fungal invasion, cucumber seeds of high vigor were inoculated with spores of Aspergillus amstelodami and stored at 85% relative humidity and 25°C for 34 days. Seeds were withdrawn daily and evaluated for the amount of internal infection by A. amstelodami and total seedborne

mycoflora, germination percentage, and moisture content. Germination decreased no more than 7% after 34 days under these relatively poor storage conditions and the majority of seeds remained uninfected. To determine whether seeds of different crop plants vary in their susceptibility to invasion by storage fungi, 6 seed lots each of 10 vegetable crops and 2 field crops were inoculated with a strain of A. flavus or A. amstelodami, and stored for 30 days at 85% relative humidity and 20-22°C. Seeds were evaluated for internal infection and total seedborne population of A. flavus and osmophilic fungi, germination percentage, and moisture content at the beginning of the experiment and after 15 and 30 days of storage. Results of this preliminary experiment/indicated that seeds of different crop plants may vary in their susceptibility to infection by storage fungi. (FC-7)

- 2. Predicting the Storability of Seeds. Accelerated aging responses and effects of chemical-soak treatment were evaluated on different seed lots of tall fescue, crimson clover, sorghum, and wheat to determine the effectiveness of each as a measure of storage potential. The seed lots of each kind responded differently to the accelerated aging tests. Some lots were reduced considerably in germination, while others were relatively unaffected. Generally, the germinative responses of tall fescue, crimson clover, and wheat seed following seeks in distilled water and solutions of ammonium chloride were quite closely correlated with those after intervals in storage. Seed lots of wheat, soybean, lettuce, and onion were ranked for storability at State College, Mississippi, by accelerated aging tests, storage under poor condition, and open storage, and for vigor at Beltsville, Maryland, by germination and seedling growth tests. Rankings for storability were not the same as rankings for vigor. Tests at Mississippi indicated that respiratory quotients might be more useful than respiratory rates for estimating relative storability. (FC-9(A
- 3. Maintaining the Viability of Seeds. Undesirable color changes in red clover were prevented over a 3-year period by maintaining a low seed moisture content (5.5-6.9%). Storing red clover seeds in glass or metal containers with CO2 reduced color change and helped prolong viability. The decline in germination of 47 kinds of seeds (from 14 families) stored over a 3- to 4-year period in three types of packages was determined. (FC-14 (E10-MQ-1(a))
- 4. <u>Biochemical Changes Associated with Seed Deterioration</u>. In storage experiments 2- to 11-year-old barley seeds were more sensitive to accelerated aging than newer seeds though they did not necessarily

differ in initial germination percentage. Oxygen uptake was variable, but the rate of CO2 evolution was greater in older seeds than in newer seeds. A 1-year wheat storage experiment involving four storage conditions was undertaken to study deterioration. no loss in viability after 6 months' storage under the three better conditions, but under the poorest condition the germination had decreased after 12 weeks. Infection with storage fungi increased from 0% after 6 weeks of storage to 66% after 26 weeks. Acceleratedaging experiments indicate that the seeds become more sensitive as the storage conditions become less favorable or the time seeds are under these conditions is increased. Rates of 0, uptake in respiration studies did not differ significantly unless a difference in viability of 20% or greater existed. The rate of CO2 evolution decreased little, if at all, even though viability was reduced. Consequently, seeds with reduced 0, uptake had higher RQ values. Seeds stored under unfavorable conditions lost their ability to utilize Glucose-14C sooner than seeds kept under better conditions. (FC-136)

### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

## A. Germination, Vigor and Dormancy

- 1. Development of methods for measuring seed vigor. Incorporation of C-14 labeled leucine into water-soluble proteins of barley and corn is roughly proportional to the vigor of the lots, even when incorporation occurs during the first two hours of imbibition. Vigor was correlated with carbohydrate-synthesizing capacity of germinating barley, wheat, oat, and rape seeds during the early hours of imbibition. results suggest that measurements of biosynthetic rates might serve as a basis for rapid and sensitive evaluation of seed vigor. relationship between changes in membrane permeability and loss of vigor in barley was investigated by (1) quantitative and qualitative determinations of sugars that leak from seeds having different levels of vigor, and (2) investigating changes in structural and biochemical properties of mitochondria isolatedffrom embryos at different stages The ratios of optical densities of certain of seed development. pairs of proteins extracted from barley and separated electrophoretically changed during the first 24 hours of germination at 25°C. Changes were markedly reduced by incubation at 2°C or by imbibing with respiratory inhibitors such as malonic acid and iodoacetate.
- 2. Methods of determining crop varieties by seed characteristics. Another set of enzymes was found useful for distinguishing between soybean varieties. These enzymes appear as a series of clear bands

on a pink background in polyacrylamide gels. The chemical reactions responsible for the visual response is the partial reduction of a tetrazolium salt, iodonitro tetrazolium violet (INT), into its formazan. This reaction does not occur in the sites occupied by the enzymes. The various components in the set of enzymes do not have the same function; for example, one of the bands demonstrated strong peroxidase activity while the others do not. Three distinct banding patterns were observed among the soybean varieties. (FC-3)

3. Environmental factors affecting germination and dormancy. A new thermogradient plate was constructed of aluminum alloy with a superior thermal-conductivity rating and a plastic cover more resistant to warping. Change-over in direction of temperature gradient was automated. Methods of presenting data visually and use of computer to assemble data in useful form were developed. Certain germination conditions induced an unusual type of abnormal germination of crambe seeds. The incidence of abnormal germination was much greater at temperatures above 28°C. The production of abnormal seedlings at lower temperatures was controlled by surrounding emerging radicles with substrata to provide uniform moisture.

Kentucky bluegrass seeds (except Var. Delta) germinate low at constant temperatures of 15-30°C. Higher temperatures prevent germination with or without a brief, promotive light treatment. Daily temperature alternations of 15-25° or 10-25°C, promote germination. The promotive effect of alternating temperatures does not depend on light and may occur in total darkness. The required high-temperature period may vary from 4 to 14 hours. Six temperature-alternation cycles following planting gave as high germination as continuous cycles throughout the germination test. Given sufficient time, the temperature alternation tended to override the inhibitory effects of long light periods. Potassium nitrate overcame the inhibitory effects of long periods of light. (FC-5)

4. X-rays and radiograms as aids in analyzing seeds. The applicability of X-ray photographic techniques to seeds of narcotic and fiber crops and to crops offering special problems was studied. The X-ray BaCl<sub>2</sub> contrast method gave values in agreement with standard germination tests for Papaver and Datura, but not for tobacco. Underdeveloped seeds in F<sub>1</sub> seed populations of paddy, okra, tomato and brinjal could be rapidly isolated by radiographic screening. Mechanical damage to the radicle and hypocotyl region of soybean showed up as hair-line fractures in X-ray radiographs. (FC-10 (A7-MQ-2(a))

- 5. Germination inhibitors in seeds. The aldehyde fractions obtained from Hordeum spontaneum and Triticum dicoccoides were analyzed. Two new aldehydes, anisaldehyde and salicylaldehyde, were identified, the latter known to be a germination inhibitor. Quantitative measurements of p-hydroxybenzoldehyde and vanillin in H. spontaneum showed a decrease of 20% for the former and 40% for the latter after 3 months of storage. (FC-11 (A10-MQ-1(a))
- 6. <u>Biochemical basis of seed germination</u>. The development of proteolytic enzyme activity in lettuce seeds was found to be under the dual control of another enzyme present in dry seeds and an inhibitor of the proteolytic enzyme. These interact so that the enzyme is released. Radioactive phytin was successfully synthesized for use in phosphate transfer studies. (FC-12 (A10-MQ-6(k))
- 7. Invisible marking of seeds for identification. The problem of marking seeds consisted of three major aspects: Methods of marking seeds properly with invisible substances, choice of marking substances, and methods of identifying the invisible substances. Wet methods, including immersion and spraying techniques, and dry methods employing marking powders were studied. Wet methods were limited to aqueous solutions of marker compounds. These had the disadvantage of taking more time and introducing the extra step of drying the seeds. However, the wet methods gave a more homogeneous covering and possibly firmer adherence to the seed. The chemicals tested and the corresponding methods of identification were: (1) Minor elements and inorganic solutions (michrochemical), (2) fluorescent substance (physical), and (3) organic acid-base indicators (chemical). Each of these kinds of substances was acceptable for certain seeds. No order of efficiency among the methods was established. Each method must be adapted to the kinds of seeds to be treated. Fluorescent dyes were especially promising because of rapid identification under ultraviolet light, but comparison with a blank is necessary with fluorescent markers. (FC-13 (A10-MQ-5(a))
- 8. Physiological maturity and dormancy of grass seeds. After 3 years of storage of Poa pratensis seeds, dormancy increased as the storage temperature was decreased. At 25°C there was over 50% dormancy in Poa after 3 years; at -20°C, dormancy was complete. There was evidence that in Dactylis glomerata storage in panicles may be preferable to storage of separated seeds. The degree of maturity at harvest did not affect storability when seeds were in panicles, whereas separated seeds which had been harvested in the early dough stage did not store well. (FC-15 (E10-MQ-3(a))

9. Determining the purity of certain grass seeds. Research was conducted to determine if auxiliary treatments could be applied to enhance the separation of pure and inert components of buffalograss (Buchloe dactyloides) seed in the flotation purity analyses. The buffalograss burs apparently contain air pockets between the outer glumes and the caryopses causing the pure seeds to have an apparent density as low as the inert seeds. Application of a vacuum of 18 inches mercury for approximately 45 seconds with the seed submerged in the flotation solution was successful in overcoming this source of error. The vacuum procedure reduced the error of purity analyses from over 30% without treatment to less than 4% with the treatment. (FC-92)

# B. Seed Pathology

- 1. Methods of detecting and identifying seedborne pathogenic fungi. Helminthosporium oryzae, the incitant of a serious seedborne disease of rice throughout the world, produces a brown pigment when grown on agar to which certain polyphenolic compounds have been added. When rice seeds were placed on agar containing either caffeic acid (0.05M) or protocatechuic acid (0.05M), areas of brown pigmentation ("halos") were produced in 4 days around fungus colonies of Helminthosporium oryzae. Isolates of Alternaria spp., Phoma spp., Fusarium spp., and Epicoccum spp. comprised the identifiable isolates from rice that did not produce halos. Isolates of Piricularia oryzae, a seedborne incitant of a very important rice disease of world-wide occurrence, and isolates of Helminthosporium spiciferum, occasionally reported from rice seeds, also produced halos. Certain other seedborne Helminthosporium spp. not found in rice seeds produced halos or clear zones. (FC-4)
- 2. Methods for testing seed for seedborne organisms. Several species of bacteria, especially Bacillus cereus var mycoides inhibit the growth of seedborne fungi and interfere with their detection. This bacterial competition can be controlled in blotter and agar tests by adding terramycin or streptomycin. In this way an increasing percentage of infected seeds was detected. Host resistance was reduced by stopping germination with 2,4-D or by killing the imbibed seeds by freezing at very low temperatures. The latter method was especially effective for the blotter tests. After deep-freezing, long-wave ultraviolet light was used to induce sporulation as an aid in fungus identification. Blotter and agar methods were compared for certain infections using the soil emergence test as a standard. method, including chlorine pretreatment of seeds was preferable for Ascochyta spp. in peas, Colletotrichum lindemuthianum in beans and Phoma betae in beets. The blotter test with deep-freezing for killing seeds has many advantages for Fusarium spp. in cereals and Septoria nodorum in wheat. (FC-16 (E19-AMS-11(a))

#### C. Protein of Feed Grains

1. Measuring protein content of grain sorghums and other feed grains. A rapid, inexpensive test for determining protein content in grains was developed. A gram of powdered cupric carbonate is added to the ground meal dispersed in a dilute alkali-alcohol solution. The mixture is shaken, filtered, and the colorimetric reading is recorded. The color intensity of the filtrate is proportional to the protein concentration. Meter readings of 400 samples of various grains representing three crop years were correlated with Kjeldahl proteins. All correlation coefficients were highly significant. The test takes about 5 minutes when samples are run sequentially. Reagent costs are about 5¢ per sample. Standard laboratory equipment is needed to perform the test. (FC-26)

# Publications - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

# Maintaining the Viability of Seeds

- Lowig, E. 1968. Einfluss von Lagerung und Verpackung auf die Erhaltung der Keimfahigkeit. SAFA-6. Jahrg. Nr. 7(S.19) Bad Godesberg, Rheinallee 4a. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1968. Hier spricht die Forderungsgemeinschaft fur Saatgutforschung. SAFA-6. Jahrg. Nr. 17/18/19. Bad Godesberg, Rheinallee 4a. (FC-14 (E10-MQ-1(a))
- Lowig, E. 1968. Untersuchung der Samen-Hartschaligkeit bei weiteren Sorten von Phaseolus vulgaris. Saatgut-Wirtschaft. Fachzeitschrift fur Samen und Saaten. (FC-14 (E10-MQ-1(a))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

# Environmental Factors Affecting Germination

- Hendricks, S. B., V. K. Toole and H. A. Borthwick. 1968. Opposing actions of light in seed germination of <u>Poa pratensis</u> and <u>Amaranthus arenicola</u>. Plant Physiology 43(12):2023-2028. (FC-5)
- Larsen, A. L. 1969. Chemical methods for variety identification. Soybean News 20(3):3. (FC-3)
- Larsen, A. L. 1969. Isoenzymes and varietal identification. Seed World 104(9):5-6. (FC-3)

# Development of a Method for Measuring Seed Vigor

- Abdul-Baki, A. 1969. Metabolism of barley seeds during early hours of germination. Plant Physiology 44(5):733-738. (FC-2)
- Mayer, A. M. and Y. Shain. 1968. Zymogen granules in enzyme liberation and activation in pea seeds. Science 162:1283-1294. (FC-12 (A10-MQ-6(k))
- Shain, Y. and A. M. Mayer. 1968. Activation of enzymes during germination: Amylopectin-1, 6-glucosidase in peas. Physiologia Plantarum 21:765-776. (FC-12 (A10-MQ-6(k))
- Woodstock, L. W. 1968. Relationships between respiration during imbibition and subsequent growth rates in germinating seeds. Third International Symposium on Quantitative Biology of Metabolism. Ed. A. Locker. pp. 136-146. (FC-2)
- Woodstock, L. W. and Combs, M. F. 1967. Application of the respiration test for seed vigor to pea. Proc. Offic. Seed Anal. 57. (FC-2)

## Methods for Detecting and Identifying Seedborne Fungi

- Kulik, M. M. 1968. A compilation of descriptions of new <u>Penicillium</u> species. USDA, Agr. Handbk. 351, 80 pp. (FC-4)
- Kulik, M. M. and R. T. Hanlin. 1968. Osmophilic strains of some Aspergillus species. Mycologia 60:961-964. (FC-4)
- Limonard, T. 1968. Ecological aspects of seed health testing. Int. Seed Test. Assoc. 631,53.011.5:632-4.093.2 (FC-16 (E19-AMS-11(a))
- Tempe, J. de. 1968. The detection of <u>Helminthosporium</u> and <u>Fusarium</u> spp. in ryegrass and meadow fescue seed samples. Proc. Int. Seed Test. Assoc. 33(3):541-542. (FC-16 (E19-AMS-11(a))
- Tempe, J. de. 1968. Some experiments on testing beet seed for Phoma betae. Proc. Int. Seed Test. Assoc. 33(4):567-572. (FC-16 (E19-AMS-11(a))
- Tempe, J. de. 1968. The quantitative evaluation of seed-borne pathogenic infection. Proc. Int. Seed Test. Assoc. 33(4):573-581. (FC-16 (E19-AMS-11(a))
- Tempe, J. de. 1968. An analysis of the laboratory testing requirements of two seed-borne diseases. Proc. Int. Seed Test. Assoc. 33(4): 583-588. (FC-16 (E19-AMS-11(a))
- Tempe, J. de. 1968. Testing oat seed for <u>Fusarium</u> spp. and <u>Helminthosporium</u> avenae. Proc. Int. Seed Test. Assoc. 33(4):589-602. (FC-16 E19-AMS-11(a))
- Tempe, J. de. 1968. The quantitative effect of light and moisture on carrot seed infections in blotter medium. Proc. Int. Seed Test. Assoc. 33(3):547-553. (FC-16 (E19-AMS-11(a))

#### AREA 5

#### LIVESTOCK AND MEAT

Research Activity Code: 2-50-707-14-204

2-50-702-14-214

2-50-501-14-217

2-50-412-14-223

# USDA and Cooperative Program

	Scientist Man-Years F.Y. 1969
Location of Intramural Work	Research Problem Area:
	412 : 501 : 707 : Total
Maryland (Beltsville)	1.0 : 5.0 : 0.5 : 6.5
Total	1.0 : 5.0 : 0.5 : 6.5

Intramural program is supplemented by extramural support, representing (a) 2.5 SMYs at State Agricultural Experiment Stations  $\frac{1}{2}$ , and (b) P.L. 480 funds in one country representing 13,550 U.S. dollars equivalent.

1/ RPA 412 1.0; RPA 501 0.5; and RPA 707 1.0

#### Progress - USDA and Cooperative Program

### RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

- 1. Postmortem Changes in Quality and Functional Properties of Bovine Muscle Proteins. Researchers at Oklahoma State University have developed improved methodology for isolating pure myosin and actin from bovine muscle. In their studies of the polymerization of G-actin, they found that optimum polymerization occurred in the 0-25.71 mM range of MgCl2 whereas neither CaCl2 nor BaCl2 had a significant effect. Both thioglycolate and cysteine maintained reduced conditions and altered the pH of the solution, thus preventing the formation of the polymer. (FC-102(A))
- 2. Yield Grades of Lambs as Related to Quality Loss During Storage and Transit. A study was initiated under a cooperative agreement with the American Sheep Producers Council to determine the amount of

shrinkage, quality change, and growth of microorganisms on chilled ovine carcasses representing various yield grade categories during shipping. The first three of a total of 12 shipments have been studied. (FC-140(A))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. Meat Flavor as a Palatability Standard. Work was initiated to determine whether a relationship exists between the glycogen structure present in meat and the meat flavor. Available analytical procedures for use in the study were modified to minimize changes occurring in glycogen. One method for the isolation of glycogen by the cold water method appears to result in retention of the natural glycogen configuration and therefore is the method of choice for isolating glycogen. (FC-55)
- 2. Objective Measurement of Beef Maturity. Data collected from analyses of muscles of Black Angus steers, 6, 12, 18, 24, 30, 36, or 48 months old, indicate a direct relationship exists between animal age and average iron content. A fraction containing three enzymes, cathepsin,

  -glucuronidase, and
  -galactosidase, was isolated from bovine
  Longissimus dorsi muscle. These enzymes were activated by membranerupturing conditions and also by simultaneous freezing and thawing.

  These results provide good evidence that lysosomes are present in bovine
  L. dorsi muscle. The experimental conditions of pH, ionic strength,
  column temperature, and nitrogen pressure needed for single column ion
  exchange chromatographic resolution of all the acidic, neutral, and
  basic amino acids commonly found in beef muscle extracts were determined.

  (FC-56)
- 3. Comparison of Quality of Beef from Bulls and Steers. Chemical analyses, histological studies and determinations of myoglobin and water holding capacity of rib samples from bulls and steers in the 9, 12, 15, and 18 month age groups have been carried out by researchers at the University of Nebraska. Taste panel evaluations have also been completed through the 15 month age group. The second replicate of the study, consisting of 144 Angus calves from two sources and separated into age-at-slaughter, sex and level of energy groups, was initiated. (FC-114(A))
- 4. Occurrence of Adipose, Connective and Fibrillar Tissues in Muscles of Slaughter Animals. A study was initiated by researchers at the University of Lublin, Poland, under a PL 480 grant to determine the composition, tenderness and nutritional value of seven muscle groups from cattle, swine, sheep and chickens representative of various ages, sex and states of nutrition. Histological, chemical and organoleptic analyses of samples from eight head of cattle have been carried out. (FC-135)

5. Biochemical Characterization of Microbial Deterioration of Meat.
Reazurin was found difficult to use for estimation of redox potential of microorganisms because of valence transition of the ferrous-ferric ions in the heme molecules. Use of 5,5'Dithiobis-(2-nitro-benzoic acid) and similar dyes may be more effective for measuring the relative oxidation-reduction potential spectrophotometrically as an index of numbers of microorganisms present and will be investigated. (FC-139)

# RPA 707 - PREVENT TRANSMISSION OF ANIMAL DISEASES AND PARASITES TO PEOPLE

- 1. Identification of Meat Slaughtered by Approved Methods. There is some evidence to indicate in normal animals that Clostridium septicum organisms from the intestines migrate directly into the muscle tissues following death. Samples collected at death and 4 hours postmortem were negative, however, when assayed for that organism using the fluorescent antibody staining technique. (FC-58(C))
- 2. Identifying Species of Origin of Heated Ground Meat. An immunochemical technique for identification of species of origin was developed by researchers at the University of Illinois. Monospecific antisera of avian, bovine, equine, ovine and porcine origin were prepared. The greatest difficulty in using the presently developed method appears to be the variability in antibody production. (FC-100(A))
- 3. Identification of Vegetable Proteins Added to Meat Products as an Aid in Inspection. Identification of isolated soy proteins by recently published methods has been investigated. Substitution of thin slab polyacrylamide gel for starch in electrophoretic development of protein fractions reveals two heavy fractions common to isolated soy protein extracts. The problem of reproducing these electrophoretic patterns lies in finding a suitable solvent for the extraction of soy protein from the homogenized meat product. Salt solutions capable of solubilizing 10X the amount of soy protein found in a meat product with 3% added soy protein do not yield the identifying protein patterns. Apparently the high fat content present in meat products prevents the complete extraction of added soy protein by salt solutions. (FC-118)

# Publications - USDA and Cooperative Program

# RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

Marriott, N. G., Naumann, H. D., Stringer, W. G. and Hedrick, H. B. 1967. Color stability of pre-packaged fresh beef as influenced by pre-display environments. Food Tech. 21:104-106. (MQ 2-75) Stringer, W. C., Bilskie, M. E. and Naumann, H. D. 1969. Microbial profiles of fresh beef. Food Tech. 23:97-102. (MQ 2-75)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Breidenstein, B. B., Cooper, C. C., Cassens, R. G., Evans, G. and Bray, R. W. 1968. Influence of marbling and maturity on the palatability of beef muscle. I. Chemical and organoleptic considerations. J. Ani. Sci. 27:1532-1541. (FC-57(C))
- Carpenter, Z. L., Smith, G. C., King, G. T., Shelton, M. and Hoke, K. E. 1969. Effects of chronological age upon palatability of lamb. Research Reports Sheep and Angora Goat, Wool and Mohair-1969. PR-2639:20-23. (FC-64(A))
- Carpenter, Z. L., Smith, G. C., King, G. T. and Hoke, K. E. 1969. Lamb carcass maturity and its relationship to palatability. Research Reports Sheep and Angora Goat, Wool and Mohair-1969. PR-2640:23-25. (FC-64(A))
- Carpenter, Z. L., Smith, G. C., King, G. T., Shelton, M. and Hoke, K. E. 1969. Relationship of chronological age to physiological maturity of ewe and wether lambs. Research Reports Sheep and Angora Goat, Wool and Mohair-1969. PR-2641:25-27. (FC-64(A))
- Cooper, C. C., Breidenstein, B. B., Cassens, R. G., Evans, G. and Bray, R. W. 1968. Influence of marbling and maturity on the palatability of beef muscle. II. Histological considerations. J. Ani. Sci. 27:1542-1546. (FC-57(C))
- Crowe, P. F. and Spicer, D. S. 1968. Peak-width equation and some related values. J. Gas Chromatog. 6:515-518. (FC-55)
- Crowe, P. F. and Spicer, D. S. 1969. Determination and application in GLC of semilog function of retention time and chain length: Effect of temperature variation on function. J. AOCS. 46:5-7. (MQ 3-62)
- Davis, C. E. and Hoke, K. E. 1969. Do your beef coolers have adequate lighting? National Provisioner 160:13-15. (FC-60)
- Hoke, K. E. and Davis, C. E. 1969. Lighting conditions for evaluation of beef carcasses. Food Tech. 23:91-93. (FC-60)

#### AREA 6

#### OILSEEDS AND PEANUTS

Research Activity Code: 1-38-702-14-202

1-38-501-14-211 1-38-408-14-212 1-38-501-14-216

# USDA and Cooperative Program

	:_	Scient	ist	Man-	Yea	rs F.	Y.	1969
Location of Intramural Work	:_	Research Problem Area:						
	_ :	408	:	501	:	702	<u>:</u>	Total
	:		:		:		:	
Georgia (Albany)	:	0.5	:	1.5	:	1.0	:	3.0
North Carolina (Raleigh)	:		:	2.0	:	1.0	:	3.0
Texas (College Station)	:		:		:	0.5	:	0.5
	:		:		:		:	
	:		:		•		:	
Total	:	0.5	:	3.5	:	2.5	:	6.5
	:		:		:		:	

Intramural program is supplemented by extramural support representing P.L. 480 funds in two countries representing 39,861 U.S. dollars equivalent.

#### Progress - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. Commercial Drying, Handling and Storage of Peanuts. Production histories of lots of farmers' stock peanuts found to contain visible Aspergillus flavus growth when marketed indicated that (1) A. flavus will grow and produce aflatoxin in peanuts prior to digging; (2) insect damage is often associated with A. flavus growth on farmers' stock peanuts; and (3) considerable field contamination of peanuts with aflatoxin may occur under extreme drought conditions. The study of the geographical distribution of production areas for peanuts found to contain A. flavus growth in North Carolina shows that these peanuts

were produced in areas that received less than 1" of rainfall during extremely hot weather from August 20 to September 21, 1968, at the time that the peanuts were approaching maturity in the soil.

Studies were made to relate off-flavor in peanuts cured at elevated temperatures to an oxygen deficit in the peanuts. Measurements with micro-oxygen probes showed that the partial pressure of oxygen in the center of immature kernels curing at 75 and 125°F and mature kernels curing at 125°F dropped to less than 125 mm, 100 mm, and 0 mm of mercury, respectively. This relationship suggests that oxygen stress may cause off-flavor production in peanuts since previous studies have shown that off-flavor is produced in immature peanuts cured at 125°F but not at 75°F and less off-flavor is produced in mature than in immature peanuts cured at 125°F. (FC-46)

2. Effects of Storage Temperatures on Quality of Vegetable Salad Oils. Flavor scores of soybean salad oils showed negligible changes during 2 years of storage between oils stored at 55, 70, and 80°F and the refrigerated oils. Oils held at 90°F for 2 years had some decrease in quality. Prior to the end of 1 year's storage there were definite decreases in flavor of oils stored at 100 and 110°F. After a 4-day heat treatment (140°F) decrease in flavor scores of oils held at 55, 70, 80, and 90°F was somewhat less than was found on oils before heat treatment.

Cottonseed salad oils stored for the 2-year period at temperatures from 55 to 110°F showed little or no decrease in quality as indicated by difference in flavor scores. However, when these oils were heat treated, exposed to air, for 4 days at 140°F there was an improvement in quality as shown by the flavor score difference. (FC-76)

3. Effects of Improved Harvesting, Handling, Conditioning and Storage on Market Quality of Peanuts. Shelled Spanish peanuts were stored under six different combinations of temperature, air flow and packaging. After 16 weeks, peanuts showed definite darkening and/or dulling. The degree of darkening was closely related to the percentage of moisture loss during storage. A general lowering of the iodine value of the peanuts was also noted and is related to the first stages of oxidative rancidity. While intermittent heated air and infrared drying at moderate temperature (below 135°F) do not reduce overall quality of peanuts, some increase in skin slippage was found at the higher drying temperatures.

Maturity appears to be a factor in the resistance of Runner and Florigiant peanuts to invasion by  $\underline{A}$ .  $\underline{flavus}$  and resultant contamination by aflatoxin. Immature Spanish peanuts do not appear to resist mold invasion to the same degree.

The flavor of windrowed peanuts is preferred to those with zero exposure and the flavor of early peanuts is preferred over late peanuts with zero exposure and 7-day random windrow; however, late peanuts are preferred over early for 7-day inverted windrow. It was found that the optical density (or maturity index of zero exposure) is significantly higher in immature peanuts in windrowed peanuts. The iodine value is higher for the zero exposure peanuts than for the windrowed peanuts. (FC-83)

4. Improved Methods for Drying Peanuts with Refrigerated Air. Generally quality of the peanuts is found to be lowered when dried with refrigerated air and peanuts in the top of the bin were of lower quality than those in the bottom of the bin. Free fatty acids of low-temperature dried peanuts, for the most part, are higher and the flavor is preferred less than that of the ambient controls. Optical densities of the oil from the treated samples were significantly higher than the ambient controls, indicating that the maturation process was delayed because of the low temperatures used in drying. (FC-107(A))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. <u>Estimating Stinkbug Damage in Soybeans</u>. The decrease in density of soybeans with stinkbug punctures is the basis for a second method (floating) to determine extent of damage. The effect of stinkbug damage on quality has been related to the resulting decrease in oil content, increase in free fatty acids, and increase in protein content. (FC-29)
- 2. Methods of Identifying and Evaluating Stinkbug Damage in Soybeans. Threshing produced more splits and broken seedcoats in stinkbug damaged soybeans than in undamaged beans. A stinkbug infested plot produced 8.4% less soybeans by weight than an uninfested adjacent plot of same size. Damaged beans had no effect on undamaged beans in the same lot during storage. Damaged beans examined by microscopic sectioning ranged from 1.3 to 51.5% damaged tissue. No increase in damaged tissue was found after storage. Results for differences in oil content, fat acidity, and protein content were somewhat inconclusive. In general, damaged soybeans contained lower oil content, higher fat acidity, and higher protein content than nondamaged beans. A few exceptions to this tendency were reported. Taste panel results indicated probable lower palatability of stinkbug damaged beans. (FC-30(C))
- 3. Objective Measurements of Market Quality in Raw Peanuts. The influence of maturity and curing temperature on the volatile profile have been studied. It has been shown that the total volatile content of the peanut reaches its maximum at 7-8 weeks after pegging, declines

to a minimum near 11 weeks and maintains values near the minimum beyond full maturity. Most individual components also follow this same pattern but pentane only drops to 50% of its maximum and constitutes between 50-60% of the total volatiles at 11 weeks and beyond. Results from studies using various curing temperatures suggest that three compounds present in peanuts, ethyl acetate, ethanol and acetaldehyde, might indicate flavor and quality deterioration due to increasing curing temperature. Taste panel results suggest that presence of any ethyl acetate in a sample indicates off-flavor and poor quality. Acetaldehyde content increased with increasing curing temperature and ethanol content increased sharply at curing temperatures of 45 and 50°C. Studies of the changes in the volatile profile of peanuts of different maturity levels cured at 50°C gave additional evidence to support the use of these compounds as quality indicators.

Changes in the activity levels of alcohol dehydrogenase and lipoxidase in peanut kernels were determined during maturation and compared to changes in the individual volatile components and possible relationships were indicated. (FC-47)

- 4. Method and Equipment for Evaluating the Quality of Peanuts. (a) Laboratory studies on the design of a diverter-type automatic sampler for shelled peanuts showed that the diverter cup should be 1.5" wide and that sampling accuracy was not affected by diverter velocities up to 45" per second. Percentages of typical types of foreign material found in the samples duplicated those in the population within 95% tolerance limits. Impact between the leading side of the diverter and the falling stream of peanuts was observed at diverter velocities over 45" per second. High-speed movies confirmed that very little impact occurred in the stream of peanuts when the diverter velocity was 30" per second. Arrangements have been made to test commercially available diverter-type samplers at two peanut shelling plants in the Southeast during 1969.
- (b) Computer data derived from theoretical models of contaminated lots of peanuts have been cataloged to provide data for the development and testing of sampling plans for aflatoxin. A type of attribute sampling plan was developed and introduced to the peanut industry. The probabilities of rejecting or accepting lots of peanuts were computed for several sampling plans considered by the peanut industry. The plan adopted by the industry for the 1969-70 season is based upon data provided by this project. Samples from contaminated lots of peanuts have been obtained for studies on the distribution of aflatoxin in shelled peanuts. The experimental data obtained will be used to test the theoretical model previously developed. (FC-48)

5. Objective Measurement of Market Quality on Raw Peanuts. A new method for measuring total acidity of peanuts was developed based on the determination of the electrical resistance of a butanol-hexylamine extract of a sample of peanuts. This measurement appears related to the presence of off-flavor components.

An average kernel weight count of a sample of peanuts appears related to quality.

Procedures for making tristimulus colorimetric measurements of peanut butter, raw peanuts, roasted peanuts and roasted-blanched peanuts were developed.

A laboratory method for measuring blanchability of peanuts was worked out.

A new technique for measuring the moisture distribution or displacement value of a sample of peanuts was worked out. This determination is related to the heat damage in peanuts. (FC-84)

6. Development of Methods and Techniques for Evaluation of Market Quality of Oilseeds and Oilseed Products. Special samples of ground soybeans have been prepared and analyzed for a cooperative effort with the Instrumentation Research Laboratory to develop a rapid method of determining moisture, oil, and protein by infrared adsorption. A significant improvement in the official method of the American Oil Chemists' Society for the determination of oil in soybeans has also been developed. (FC-150)

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

1. Rapid Detection of Molds and/or Aflatoxin in Peanuts. The visible Aspergillus flavus method for detecting lots of farmers' stock peanuts contaminated with aflatoxin was evaluated with the following findings: (1) 364 composite samples containing all kernels from grade samples taken from 3,640 lots of Seg. 3 peanuts contained an average of 203 p.p.b. aflatoxin; (2) aflatoxin was detected in 99% of samples from Seg. 3 peanuts while approximately 38% of samples from Seg. 1 peanuts contained some aflatoxin; (3) inspectors correctly identified A. flavus on kernels from Seg. 3 peanuts 94% of the time; and (4) farmers' stock peanuts should be placed in Seg. 3 storage even though A. flavus is found only on small shriveled kernels in the LSK.

Grade samples were analyzed from 1,434 lots placed in Seg. 1 storage and 327 lots placed in Seg. 3 storage at 27 buying points in Georgia and Alabama. The samples placed in Seg. 3 storage had an average of 337 p.p.b. aflatoxin compared to an average of 18 p.p.b. in those placed in Seg. 1 storage.

As requested by the Peanut Administrative Committee, recommendations have been developed for improvement of the visible  $\underline{A}$ .  $\underline{flavus}$  method during the 1969 marketing season. A study of 305 samples of Seg. 3 peanuts showed that the incidence of  $\underline{A}$ .  $\underline{flavus}$  kernels was as high in the LSK portion of the samples as in the remainder of the sample. The Peanut Administrative Committee has recommended that LSK from 1,800-gram samples be inspected for visible  $\underline{A}$ .  $\underline{flavus}$  growth during the 1969 marketing season. (FC-49)

2. Physiological and Biochemical Factors in the Production of Aflatoxin by Aspergillus flavus. Toxic Aspergillus flavus strains maintained on media containing yeast extract or groundnut cake or both showed increased yields of aflatoxins upon culturing.

Tests with eight strains of  $\underline{A}$ .  $\underline{flavus}$  isolated in India showed six strains produced only B, one strain produced small amounts of B and larger amounts of a higher  $R_f$  fluorescent compound and one strain produced no aflatoxin.

Possible intermediates in aflatoxin biosynthesis and compounds related to aflatoxins in  $\underline{A}$ .  $\underline{flavus}$  have been isolated and classed into two groups based on  $\underline{TLC}$  behavior, ultraviolet spectra, and solubility in chloroform and petroleum ether. A cell-free system to study aflatoxin biosynthesis has been prepared and the incorporation of acetate-l- $^{14}$ C into aflatoxin was shown to be associated with the mitochondrial fraction. The incorporation of  $^{14}$ C-leucine and  $^{14}$ C-mevalonate into aflatoxin was also investigated using the cell-free system.

Studies of the incorporation of acetate-1- $^{14}$ C into lipid and aflatoxins by A. flavus mycelium indicated that phospholipids contained the greatest amount of incorporated activity. Among the phospholipids, phosphatidyl inositol had the highest activity and among the neutral lipids, free fatty acids were the most radioactive. The amount of radioactivity incorporated into aflatoxins was comparable with the amount incorporated into the lipid fraction. (FC-54 (A7-MQ-7(a))

3. <u>Development and Control of Mycotoxins in Spanish Peanut</u>. Significant or consistent aflatoxin contamination was not found associated with fungicide treatments or with regular vs. inverted windrows in peanuts

from experimental plots in south Texas in 1968. Studies of successive single-spore generations showed that some isolates of the A. flavus group are quite variable in the ability to produce the aflatoxins. Variability was also found in morphological characteristics. In contrast, such variability could not be demonstrated in other strains. The aflatoxins were found to be most stable in the slightly acid to neutral range and most liable to decomposition by a strong base. No significant difference in the aflatoxin concentration was found to be associated with fluorescent or nonfluorescent kernels separated from commercial "pickouts." The number and kinds of fungi isolated from the two fractions also did not vary greatly between fractions. (FC-78)

4. Rapid Detection of Molds and/or Fungal Metabolites. The millicolumn chromatography method for aflatoxin was simplified and time for assay reduced to about 10 minutes (the millicolumn tubes are now being produced commercially). A procedure for quantifying the millicolumn has been developed, based on removing the silica gel containing the aflatoxin band from the millicolumn, washing it with diethyl ether, and extracting with methanol. The methanol is removed with a flash evaporator and  $\frac{1}{2}$  ml of a 3% acetonitrile in benzene solution is added. Three hundred  $\mu l$  of this is plated on a 0.75 mm thin-layer plate and developed. The aflatoxin spots are then quantified on a fluoromicrophotometer.

A simple procedure for determining the aflatoxin-producing potential of fungi was developed. It is based on the detection and quantification of aflatoxin by means of millicolumn chromatography and TLC in a chloroform extract of the fungi grown under standard conditions. (FC-85)

5. Mechanisms of Fungal Deterioration Affecting Market Quality of Peanuts. A method has been developed for quantitative analyses of changes in levels of various amino acids during invasion of peanuts by fungi. Amino acids are converted to their N-acetyl-N-butyl esters and separated by gas-liquid chromatography on column coated with methyl silicone.

Green peanuts were found to be highly resistant to mold invasion. After being dried and rehydrated, this defense mechanism was destroyed. (FC-120)

6. <u>Biochemical Characterization of Fungal Metabolites</u>. A highly fluorescent metabolites extracted from a culture of <u>Penicillium urticae</u> was purified and identified as griseofulvin by means of UV,

IR, NMR, mass spectra, TLC and GLC. This compound has a potent fungistatic action, particularly against those fungi responsible for dermatomycosis in man and animals. Two minor components were also found in the extract. Mass spectrum and NMR spectrum of one of these compounds showed it to be dechlorogriseofulvin. (FC-133)

7. Biology of the Fungus Aspergillus Flavus Link and its Infectivity to Plants and Animals. The general mycoflora of the kernels of peanut varieties grown in Israel were similar but penetration of Aspergillus flavus into kernels was found to vary considerably among varieties. The incidence of the fungus was always low in the Virginia S.M. variety. However, potentially pathogenic fungi, i.e., A. niger and Fusarium solani, penetrated kernels effectively. Although A. flavus was widespread in Israel and the population contained an extremely high prevalence of toxigenic isolates, a marked contamination of the peanut crop did not necessarily follow. Cultural and humidity factors in the field and in storage had a marked effect. In addition, the prevalence of fungi antagonistic to A. flavus must be taken into account.

Application of aflatoxin to the skin of rabbits was found to cause not only profound histological changes but induced changes in the inner organs (especially the liver) and eventually death. (A10-CR-46)

## Publications - USDA and Cooperative Program

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- Holaday, C. E. 1968. Rapid method for detecting aflatoxins in peanuts. J. American Oil Chemists' Society 45(10):680-682. (FC-85)
- Joffe, A. Z. 1968. The effect of soil inoculation with <u>Aspergillus</u> <u>flavus</u> on the mycoflora of groundnut soil, rhizosphere and geocarposphere. Mycologia 60:908-914. (AlO-CR-46)
- Joffe, A. Z. 1968. Mycoflora of surface-sterilized groundnut kernels. Plant Disease Rep. 52:608-611. (A10-CR-46)
- Joffe, A. Z. and N. Lisker. 1968. Effect of soil fungicide on development of fungi in soil and on kernels of groundnut. Plant Disease Rep. 52:718-721. (A10-CR-46)
- Schroeder, H. W. 1968. Metabolite of <u>Macrophomina phaseoli</u> that can confuse thin-layer chromatographic identification of aflatoxin B<sub>2</sub>. Appl. Microbiol. 16:946-947. (FC-78)
- Schroeder, H. W. and Hugo Hein, Jr. 1968. Effects of diurnaltemperature cycles on the production of aflatoxin. Appl. Microbiol. 16:988-990. (FC-78)

- Welty, R. E. and W. E. Cooper. 1966. The influence of landplaster and fungicides on fungi isolated from peanut, <u>Arachis hypogaea</u>. Phytopathology 56:905 (Abstract).
- Welty, R. E. and W. E. Cooper. 1967. Effects of moisture content and time on fungi isolated from peanut <u>Arachis hypogaea</u> kernels. Phytopathology 57:464 (Abstract).
- Welty, R. E. and W. E. Cooper. 1968. Prevalence and development of storage fungi in peanut (<u>Arachis hypogaea</u>) seed. Mycopathologia et Mycologia Applicata 35:290-296.
- Dickens, J. W. and R. E. Welty. 1968. Detecting farmers' stock peanuts containing aflatoxin by examination for visible growth of <u>Aspergillus flavus</u>. Proceedings Mycotoxin Research Seminar, Washington, D. C. (June 1967), USDA. (FC-49)

#### AREA 7

#### COTTON AND COTTONSEED

Research Activity Code: 1-38-702-14-202

1-16-501-14-208 1-38-501-14-211 1-38-408-14-212 1-38-501-14-216

#### USDA and Cooperative Program

	:_	Scient	ist	Man-	Yea	rs F.	Y. 1	969
Location of Intramural Work	:	Resea	rch	Prob	1em	Area	::	
	:	408	:	501	:	702	:	Total
	:		:		:		-:	
South Carolina (Clemson)	:		:	7.0	:		:	7.0
	:		:		:		:	
Washington, D. C.	:	1.0	:	1.0	:	1.0	:	3.0
	:		:_		:		:_	
	:		:		:		:	
Total	:	1.0	:	8.0	:	1.0	:	10.0
			:		:		:	

Intramural program is supplemented by extramural support representing (a) 0.8 SMY at State Agricultural Experiment Stations  $\frac{1}{2}$ , and (b) P.L. 480 funds in one country representing 5,741 U.S. dollars equivalent.  $\frac{2}{2}$ .

# <u>2</u>/ RPA 702 0.2

# Progress - USDA and Cooperative Program

# RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

1. <u>Improvement of Official Cottonseed Standards</u>. The American Oil Chemists' Society has officially adopted the modified neutral oil method developed by the Field Crops and Animal Products Research Branch. (The method is equally adaptable for use with cottonseed and other vegetable oils.) This modified method includes use of a newly designed chromatographic assembly which converts the manual operation into a semiautomatic procedure. A patent on the design of the apparatus has been applied for

<sup>1/</sup> RPA 501 0.8

by the USDA. Since "Methods of the American Oil Chemists' Society" are used as official methods by the National Soybean Processors Association, this method will also be used in determining the quality of crude soybean oil in commercial trading.

A manuscript "Improved factors for grading cottonseed" (by M. E. Whitten, MQRD, ARS; W. T. Coleman, Paymaster Oil Mills, Abilene, Texas; and R. T. Doughtie, Jr., Cotton Division, C&MS) has been prepared for publication as a Marketing Research Report. (FC-79)

2. Effect of Cottonseed Storage on Oil Quality. It was established that there are two main gossypol-containing pigments in refined cottonseed oil. One is probably pheophytin-gossypol which can be removed, preferably, by treatment with bleaching earth. The other pigment is probably oxidized gossypol which has been removed by treatment with bleaching earth at 115-120°C, followed by treatment with alkali. (FC-128)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Effects of Previous History of Cotton Lint on Market Quality. Removal of 8% comber noils greatly improved yarn quality and spinning efficiency over that obtained from carded yarns. When comber noil removal was increased above 8% to a high of 18%, only small improvement in yarn quality and spinning efficiency resulted. The tensor opening required for optimum spinning efficiency is the same for both carded and combed yarn. The tensor opening would be different for cotton of other lengths.

Cotton harvested by spindle pickers produced better fiber and yarn quality with improved spinning efficiency than cotton harvested by strippers. Cotton harvested before frost produced better fiber and spinning qualities than cotton harvested after frost.

Fiber and yarn quality from three high quality cottons grown in the Southeast were not affected by ginning conditions except when two lint cleaners were used. The two lint-cleaner condition had a statistically significant effect in reducing fiber length and yarn quality; however, the reduction was small.

Results from the Fiber Properties Study indicate that fiber properties, particularly fiber length, are the major factors dictating the setting of the spinning frame's fiber control system for obtaining optimum spinning performance. (FC-65)

2. Methods of Measuring the Frictional Properties of Cotton Fibers. A new Spin-Draft Tester has been developed which can measure drafting force and drafting-force variability under normal drafting operations.

Good progress is being made in determining drafting-force optimums for cotton spinning. Preliminary tests involving the Spin-Draft Tester and large-scale spinning indicate that 40s yarn spun from 1.00 hank roving has optimum spinning efficiency at a drafting tenacity of approximately 18 grams. (FC-66)

- 3. Chemical Investigation of Surface Contamination of Cotton Fibers. Arsenic acid is widely used as a desiccant to prepare mature cotton plants for harvesting by stripping. Workers have developed severe rashes after handling cotton from fields which had been desiccated with arsenic acid. Since no satisfactory method was available, a rapid method for both qualitative and quantitative detection of arsenic residues on cotton was developed. (FC-67)
- 4. Cotton Fiber Testing Instruments and Techniques. Test results from the automated Fibronaire agree well with those from the manual model. The three Model 270 Production Fibrographs operated within a range of 0.01" for each span length, but mechanical means and a cotton beard must be used for setting level of results and changes in instruments or techniques require reestablishment of level. Fibrosamplers are not interchangeable but beards from the automated Fibrosampler are as good as those from the manual type with brush. The first production-line fiber-testing system results explained significantly more variation in break factor than did the present quality factors of grade, staple, and micronaire. Two lines of this system were set up by the Market Quality Research Division and the Cotton Division to determine tests per man-hour, downtime, arrangement of instruments, reproducibility, and relation to spinning. Results compared favorably with those from presently accepted instruments. All the production-type instruments for measuring fiber properties have both readout and printout systems. Two Length-Strength Analyzers (air) perform well and their measurements agree, but are not significantly better than those obtained from the capacitance type. Strength measurements are not as good as 1/8" Pressley for explaining break factor and corrections of data for micronaire effects are required. Measurements from Length Analyzer (air) are no better than those obtained from Length Analyzer (capacitance), and neither is as good as those from the Fibrograph. (FC-68)
- 5. Methodology Studies for Development of Improved Cotton Spinning
  Performance Tests. A regression equation for determining the tensor
  opening which is required for optimum spinning performance for a Duo-Roth
  drafting system has been developed. This equation, which applies only
  tocour Pilot Plant operations and to 40s yarn, uses the 2.5% span length,
  length uniformity ratio, and micronaire reading for predicting the tensor

opening. This formula has proved to be quite accurate in our work in predicting the tensor opening for optimum spinning performance for 40s yarn. Further development of this formula for use on other yarn counts is underway. (FC-70)

6. Measuring Factors Affecting Spinning Performance and Product Quality of Cotton. Evaluation of the Uster Evenness Tester and Imperfection Indicator shows that the measurements indicated by the instruments are not affected by the testing speed as long as the linear operating range of the instrument is not exceeded. The limits of this range have been defined and a mathematic model for simulating the dynamics of the spinning balloon developed. Computer programs have been written and the mathematical model yields values for the displacement and tension of the yarn for certain specific types of yarn motions.

Acoustic and electronic equipment for the detection of individual fiber breakage during the strength testing of a bundle of fibers has been developed. Upon breaking, cotton fibers generated acoustic pressure pulses of varying magnitude. Present research efforts are directed toward understanding this phenomenon.

A mathematical relationship appears to exist between the spinning tensor opening, which gives the maximum spinning efficiency, and the fiber properties of the cotton. Expressions were also developed relating the drafting force at various tensor openings to the fiber properties. The use of the new Spin-Draft Tester for measuring the drafting force at the tensor setting being used during yarn production was shown to be a useful monitoring technique for detecting changes in the properties of the roving. (FC-71(A))

- 7. Apparatus for Blending a Sample of Cotton Lint. A miniature card with separate variable speed drives for licker-in, cylinder, and doffer has been tried for blending a sample of ginned lint for fiber tests. While the approach produces an excellent card web, the blending which takes place in the machine is minor. In order to get a blend, the sample must be mixed before it is fed to the card. Mixing and carding a test sample is much too time-consuming for use in a marketing system and this approach is being discontinued. A promising approach is being investigated which involves improving the present Fibrosampler method of preparing a test specimen from a commercially drawn cotton sample. (FC-72)
- 8. Measurement of Drafting Force and Its Variability Under Cotton
  Processing Operations. Improvements have been made in the Spin-Draft
  Tester involving both the instrument which measures the drafting force
  and the electronic system which computes the data. Three types of

double apron drafting systems have been used in this work. The three systems can be set to obtain the same drafting force during drafting. Drafting force is found highly correlated with fiber length characteristics for any specific tensor opening and changing of tensor opening can affect the drafting force due to fiber length. Tensor opening, however, can be set to obtain the optimum drafting force for optimum spinning efficiency for any fiber length. The drafting force change which occurs when the direction of roving feed is reversed seems to be an efficient measurement of the major to minor hook relationship. (FC-101)

9. Effect of Cotton Properties and Residues on Fiber Quality and Grade. In recent years, cationic surfactants have been applied to seed cotton at the gin as fiber conditioners. A method for detecting trace quantities of certain cationic additives on cotton has been developed.

Complaints occurred in the Southeast during the past ginning season concerning darkening of cotton under jute bagging. Buyers were reluctant to buy cotton in areas where this condition was reported. We examined a series of cottons that were characterized by severe darkening of the lint near the bale surface under the jute bagging. These bales had been rained on while standing in gin yards, and ginners speculated that the jute bagging had contaminated the cotton. Our tests revealed that due to weathering (1) oil had transferred from the jute bagging to the cotton; (2) some water soluble products had been removed from the cotton near the surface of the bale; and (3) microbiological damage (cavitoma) had occurred. The suggested remedy was to store cotton in sheltered areas away from weather. (FC-121)

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

1. Aflatoxin Production in Cottonseed. Cottonseed, heavily infested with stored-products insects after prolonged storage in commercial facilities, was found to contain a high percentage (75%) of seed from which fungi could not be isolated after acid-delinting and surface sterilization. Only 1.3% of the seed contained Aspergillus flavus spp. However, extracts of all subsamples contained many compounds not found in sound seed with aflatoxin concentrations ranging from 0 to about 400 p.p.b./sample. None of the A. flavus isolates from these seed were able to produce aflatoxins under experimental conditions. The data indicates that aflatoxin contamination in this particular lot of seed was not associated with the insect infestation but probably occurred prior to such infection.

Glandless and glanded cottonseed were shown to be equally effective substrates for the production of aflatoxin by several toxigenic strains of  $\underline{A}$ . flavus. A strain of  $\underline{A}$ . ochraceus was isolated from cottonseed and shown by thin-layer chromatography and bioassay to be capable of producing ochratoxin. (FC-77)

- 2. Aflatoxins in Cottonseed. Findings of an initial survey indicate that aflatoxin contamination of cottonseed appears to be a minor problem in some regions of India. In an examination of fungal flora, most predominant species found among Aspergilli were A. niger, A. glaucus, A. nidulane and A. flavus. Samples of cottonseed with higher levels of A. flavus and aflatoxins showed a greenish-yellow fluorescence under ultraviolet light. (FC-97 (A7-MQ-17(k))
- 3. Effect of Ambient Storage Conditions on Molds and Aflatoxins in Cottonseed. Study of a damaged lot of cottonseed heavily contaminated with aflatoxins was conducted cooperatively with Cotton and Cordage Fibers Research Branch, Crops Research Division. Preliminary data indicate no increase in aflatoxins during storage when the aflatoxin content is already high.

An improved method of analysis for aflatoxins has been developed. The new method removes interfering pigments in cottonseed extracts, also eliminates the need for chromatographic purification of the cottonseed extracts through silica gel columns and the time and cost of aflatoxin determination is reduced by more than one-half. The results are also more reliable since degradation of the most toxic fraction, B1, has been observed when extracts from stored cottonseed are passed through silica gel columns. (FC-80)

4. <u>Effect of Commercial Storage Practices on Molds and Aflatoxins in Cottonseed</u>. This is a new project initiated on April 16, 1969. No progress to report at this time. (FC-144)

# Publications - USDA and Cooperative Program

RPA 408 - QUALITY MAINTENANCE IN MARKETING FIELD CROPS

Velasco, James. 1969. Modification of AOCS chromatographic procedure for neutral oil determination. J. Amer. Oil Chem. Soc. 46(5): 252-255. (FC-79)

#### RPA 501 - IMPROVEMENT OF GRADE AND STANDARDS

- Chapman, W. E., R. A. Mullikin'and P. E. LaFerney. 1968. Effects of gin cleaning on fiber properties and spinning quality, Pima cotton, 1965-66, with and without crusher rolls. USDA, MRR 806. (FC-65)
- Graham, John S. and Samuel T. Burley, Jr. 1968. Effect of drafting parameters on drafting force in cotton spinning. Textile Bulletin 94: 18-22. (FC-70)
- Griffin, A. Clyde, Preston LaFerney and E. H. Shanklin. 1969. Split vs. series lint cleaning. Summary Proceedings, 1969 Cotton Quality and Processing Conference, pp. 3-5. (FC-65)

- Newton, Franklin E., Samuel T. Burley, Jr. and Preston E. LaFerney. 1969. New approach to fiber quality evaluation. Summary Proceedings, 1969 Cotton Quality and Processing Conference, pp. 6-8. (FC-68)
- Perkins, Henry H., Jr. 1968. Quick check for arsenic on cotton. Textile Industries 132:151-153. (FC-67)
- Perkins, Henry H., Jr. 1969. Detection of cationic additives on lint cotton. Textile Industries 133:148-151. (FC-121)

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- Velasco, James. 1968. A simplified procedure for the determination of aflatoxin  $B_1$  in cottonseed meals. J. Amer. Oil Chem. Soc. 46(2): 105-107. (FC-80)
- Whitten, M. E. 1968. Screening cottonseed for aflatoxins. J. Amer. Oil Chem. Soc. 46(1):39-40. (FC-80)

#### AREA 8

#### WOOL AND MOHAIR

Research Activity Code: 2-43-501-14-222

# USDA and Cooperative Program

	:	Scientis	t Man-	Years F.Y	. 196	9
Location of Intramural Work	:	: Research Problem Area :				
	:	412	:	501	:	Total
	:		:		:	
Maryland (Beltsville)	:	0.1	:	0.1	:	0.2
	:		:		:	
	:		:		:	
Total	:	0.1	:	0.1	:	0.2
	:		:		:	

Intramural program is supplemented by extramural support representing (a) 0.7 SMY at State Agricultural Experiment Stations  $\frac{1}{2}$ , and (b) P.L. 480 funds in one country representing 5,100 U.S. dollars equivalent.

#### 1/ RPA 501 0.7

#### Progress - USDA and Cooperative Program

# RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

1. Objective Measurement and Evaluation of Quality Canary Yellow Coloration of Raw Wool. Three components of the canary yellow pigment were partially characterized with respect to molecular weights. The compound formed by the apparent combination of the staining compound with the free amino group of the protein resisted bleaching but sunlight caused fading. Physical properties of stained and unstained wools were similar except the stained fiber did not retain a permanent set as well as the unstained fiber. This study was completed. (FC-63 (A7-AMS-12(a))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Fineness and Softness as Quality Factors of Wool. Researchers at the University of Wyoming have invented a staple length recorder. They have also modified the Harris Research Laboratory's cutting instrument

so that a larger wool sample can be cut and a more rapid, steady force can be applied to cut wool samples. Though it is believed that fineness and softness of wool are highly related, studies of wool produced in the Wyoming area showed no definite relationship between softness index and fiber diameter. Similar information on clips obtained from the other major wool-producing areas is presently being obtained. (FC-62(C))

## Publications - USDA and Cooperative Program

#### RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

- Gupta, K. 1967. Spectrophotometric method for determining the degree of yellowness and degree of redness of yellow solutions only. Text. Res. J. 37:310-312. (FC-63) (A7-AMS-12(a))
- Gupta, K. and Bhan, M. M. 1968. Colouring matters in canary stained raw wools. Wool and Woolens of India, February 1968. (FC-63) (A7-AMS-12(a))
- Gupta, K., Patnaik, B. K. and Chipalkatti, V. B. 1968. Isolation of coloring matter from canary-colored wool. Text. Res. J. 38:107-108. (FC-63) (A7-AMS-12(a))
- Gupta, K. and Sule, A. D. 1967. Role of free sulphur & lanthionine in canary colouration of raw wools. Indian J. of Tech. 5:359-361. (FC-63) (A7-AMS-12(a))
- Juneja, K. K., Sule, A. D. and Chipalkatti, V. B. 1968. Estimation of tryptophan content of wool by barium hydroxide (baryta) hydrolysis method. Text. Res. J. 38:461-466. (FC-63) (A7-AMS-12(a))

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

Buras, E. M. Jr., Penoyer, J. A. Sr. and Hoke, K. E. 1968. Preparation of specimens from wool top and cores for electronic wool fineness determination. Text. Res. J. 38:1164-1173. (MQ 3-69(C))

#### AREA 10

#### POULTRY PRODUCTS

Research Activity Code: 2-50-707-14-204

2-50-702-14-214 2-39-412-14-218 2-39-501-14-219

# USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1969								
Location of Intramural Work	: Rese								
	: 412	: 501	: 702	: 707	: Total				
Maryland (Beltsville)	1.0		: : 1.0						
Georgia (Athens)		:	: 0.5	: 0.5	: 1.0				
Total	: : 1.0	: : 0.5	: : 1.5 :	: : 2.0	: : 5.0				

Intramural program is supplemented by extramural support representing 0.5 SMY at State Agricultural Experiment Stations  $\frac{1}{2}$ .

# 1/ RPA 702 0.2; RPA 707 0.3

#### Progress - USDA and Cooperative Program

### RPA 412 - QUALITY MAINTENANCE IN MARKETING ANIMAL PRODUCTS

1. Improved Packing, Shipping and Storage Methods for Unfrozen Poultry. Evaluations of experimental shipping boxes using dry ice as coolant were continued. Lowering carcass temperature from 4°C to 2°C before packing did not significantly lower the amount of weight loss during storage at 5°C of chickens in boxes with dry ice as coolant. By 10 days of storage, an off-odor not typical of spoilage odor was found on carcasses in polystyrene boxes with high CO2 content. This off-odor occurred despite lower total microbial counts than found in other box types. The amount or type of coolant used in fiberboard boxes at the processing plant had a negligible effect on subsequent cutting-up loss or weight loss during retail holding of chicken cut-up and film packaged at the retail level. This work was carried out in cooperation with the Transportation and Facilities Research Division, ARS. (FC-132)

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

- 1. Reducing Salmonellae Contamination of Broiler Chicken Carcasses During Processing. The effect of chlorine dioxide added to chill water on the microbiological conditions of chicken broilers was evaluated in six trials in a poultry processing plant. Broiler carcasses cooled for 45 minutes in a continuous chiller containing 6-8 p.p.m. chlorine dioxide did not exhibit significantly lower total bacterial counts than carcasses chilled without chlorine dioxide. Neither the frequency of salmonellae-positive birds nor the most probable number of Salmonella per positive bird appeared to be affected by the presence of chlorine dioxide. Most probable numbers of salmonellae per salmonellae-positive bird ranged from about 8-70, with the majority (93%) being less than 30. (FC-39; FC-41(A))
- 2. Detection of Salmonella in Poultry and Egg Products. Studies to improve and to determine the range of application of the two recently developed Salmonella detection systems were continued. The addition of 12.5 or 25.0 mg/l of brilliant green to the mannitol purple agar inhibited fermentation of mannitol by Escheria coli in the inverted tube system. The addition of 20.0 mg/l of brilliant green to semisolid tryptic soy agar was effective in inhibiting growth and/or motility of  $\underline{E}$ .  $\underline{coli}$ , Proteus vulgaris, and Pseudomonas aeruginosa in the 3-side arm glass apparatus (Banwart flask). The efficiency of detecting Salmonellanegative and Salmonella-positive samples using both techniques is obviously enhanced by the use of brilliant green.

Approximately 1,000 samples of various poultry and egg products and feeds have been analyzed with these two  $\underline{Salmone11a}$  detection systems. Employing the Banwart flask procedure, more than 99% of the confirmed  $\underline{Salmone11a}$  negative samples were detected within 48 hours. (FC-40)

# RPA 707 - PREVENT TRANSMISSION OF ANIMAL DISEASES AND PARASITES TO PEOPLE

1. Erysipelothrix Infection in Turkeys. The factor(s) responsible for, or definitely related to, the production of the carrier state of Erysipelothrix insidiosa infection in turkeys has not been identified. Factors studied were route of challenge, number of organisms in challenge dose, vaccination, type of vaccine, environmental factors (type of litter, contaminated environment, etc.) age and sex of the turkey. The only consistent finding was that if the number of organisms per ml of blood did not reach a level greater than 100,000 the organisms was either eliminated or the carrier state resulted. Thus immunization or antibiotic therapy, while of value in reducing mortality, could contribute to the production of the carrier state. (FC-43(A))

2. Sanitary Quality and Wholesomeness of Further-Processed Poultry Products. Bacteriological examinations of Eastern-type cooked turkey rolls were completed and a report was presented at the 69th Annual Meeting of the American Society for Microbiology. A manuscript has been prepared for publication in Poultry Science.

Thermal death time studies of <u>Streptococcus</u> <u>faecalis</u> in liquid egg products were initiated. Preliminary results indicate that "D" values for this organism in liquid whole egg heated at 60°C is about 3.5 minutes. (FC-38)

3. Improving Microbiological Condition of Eviscerated Broilers. Field trials to evaluate the effectiveness of chlorine as hypochlorite in the final washer on the bacteriological condition of eviscerated fryers were conducted. Final spray washing of birds with water containing 40-50 p.p.m. chlorine, prior to chilling, reduced skin surface microbial counts by about half a log over that accomplished by spray washing without chlorine. No additional benefit was derived by employing concentrations of chlorine as high as 230 p.p.m. Washing birds with concentrations of chlorine less than 40 p.p.m. was not as effective in reducing bacterial counts. Neither spray washing of birds with 29, 41, 81, or 118 p.p.m. chlorine dioxide nor chilling of birds in 6-8 p.p.m. of this agent had any significant effect on reduction of skin surface bacterial counts. (FC-122)

# Publications - USDA and Cooperative Program

- RPA 702 PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
  AND NATURALLY OCCURRING TOXINS
- Banwart, G. J. 1968. Glassware apparatus for determining motile bacteria. I. Salmonella. Poultry Science 47(4):1209-1212. (FC-40)
- Banwart, G. J. 1968. Rapid methods to screen foods for the absence of salmonellae. (Abstract). 82nd Annual Meeting Association of Official Analytical Chemists., Washington, D. C. (FC-40)
- Banwart, G. J. 1969. Rapid screening methods for detection of enteric pathogens. Round Table on the Enterobacteriaceae. Presented at the 69th Annual Meeting of the American Society for Microbiology., Miami Beach, Florida (May 1969). (FC-40)
- Banwart, G. J. and Madeleine Kreitzer. 1969. Further studies on the screening technique for determining <u>Salmonella</u>-negative samples of pasteurized dried egg. Poultry Science 48(1):235-240. (FC-40)

- Bryan, F. L., J. C. Ayres and A. A. Kraft. 1968. Destruction of salmonellae and indicator organisms during thermal processing of turkey rolls. Poultry Science 47(6):1966-1978. (FC-42(C))
- Kinner, J. A., A. W. Kotula and A. J. Mercuri. 1968. Microbiological examination of ingredients of Eastern-type turkey rolls. Poultry Science 47(5):1442-1447. (FC-38)
- Mercuri, A. J. 1968. Microbiology of further-processed poultry products. Conference on Advances in Poultry and Egg Processing, Pa. State University (October 1968). (FC-38)
- Mercuri, A. J., G. J. Banwart, J. A. Kinner and A. Sessoms. 1969.

  Bacteriological examination of cooked (foil-wrapped, oven-roasted)
  turkey rolls. Bact. Proc., p. 13. (FC-38)

#### AREA 11

#### TOBACCO

Research Activity Code: 1-38-702-14-203

1-38-408-14-213

#### USDA and Cooperative Program

		an-Years F.Y.	1969
Location of Intramural Work	Research P	roblem Area : 702	: Total
North Carolina (Raleigh)	0.2	1.0	1.2
Total	0.2	1.0	1.2

Intramural program is supplemented by extramural support representing 4.3 SMYs at State Agricultural Experiment Stations  $\frac{1}{2}$ .

# 1/ RPA 702 4.3

#### Progress - USDA and Cooperative Program

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Rapid Method for Measurement of Moisture in Cured Tobacco. The moisture meter developed under this project was tested on a tobacco warehouse floor in the fall of 1968. Samples (consistency of ten sections of lamina 2"x2" square) were taken from piles of tobacco as they were marketed. Each section was read on the moisture meter and the ten sections were composited for gravimetric determinations of moisture content. Moisture content values determined by the two methods were paired. A standard deviation from regression of ± 2.31% was achieved for samples in a range from 10-40% moisture. Precision of the instrument was found to be ± 0.30% for repeated readings on the same sample. Further research is needed to improve the accuracy of the meter. (FC-50(A))

# RPA 702 - PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS AND NATURALLY OCCURRING TOXINS

1. Investigations on the Role of Microbial Activity in Producing Carcinogens in Marketed Tobacco. A total of 6,670 microorganisms, representing more than 38 genera of fungi and various types of yeasts

and bacteria, were isolated and identified from various grades of marketed aged burley and dark air-cured tobacco stored in different tobacco producing areas. Microorganisms were Aspergillus (39.48%), Penicillium (15.21%), yeasts (15.44%), bacteria (15.74%), and miscellaneous fungi and actinomycetes (14.14%). A. flavus was the largest species isolated (29.18%). The next most frequent isolated species were A. niger, A. clavatus, A. giganteus, P. frequentans, and P. notatum. In general, there were fewer fungi associated with the dark aif-cured aged tobacco than with the burley tobacco. Only slightly higher numbers of microorganisms were isolated from the "moldy" (damaged) tobacco than from "sound" (nondamaged) tobacco. More microorganisms were found to associate with the flyings, lugs or cutters, and tips than the other grades of tobacco. Ninety percent of the  $\underline{A}$ .  $\underline{flavus}$  isolates tested were found to possess aflatoxin-producing potentials. The recovery of aflatoxins from artificially infested, environmentally-grown tobacco at various curing stages was confirmed experimentally. This completes the contracted research. (FC-52(C))

- 2. <u>Microorganisms of Burley Tobacco and Their Effect on Its Chemical</u>
  <u>Composition</u>. No work done on this project since the last report. (FC-53(C))
- 3. Investigations on the Carcinogens and Other Toxic Metabolites Produced by Tobacco Fungal Flora. A total of 9,659 fungi, representing 20 genera, were identified from 53 USDA grades of warehouse dark air-cured and burley tobacco. Of the total, 28.2% were from 38 grades of dark air-cured and 71.8% were from six varieties of burley. Aspergillus, Cladosporium, Penicillium, and Alternaria were the predominant fungi isolated. More Aspergillus and Penicillium were found on dark air-cured and more Cladosporium and Alternaria were found on burley.

Aflatoxins  $B_1$  and  $G_1$  were extracted from <u>A</u>. <u>flavus</u> isolates, purified and characterized by UV and infrared spectrophotometry and melting point determinations.

Several metabolites, other than aflatoxins, were extracted from 18 isolates of fungi (Aspergillus, Penicillium, Cladosporium, Acladium, Gliometix, and Spicaria) and found to be toxic to four bacteria, Bacillus megaterium, Bacillus brevieus, Bacillus sp X-1, and Bacillus sp.

Kojic acid was extracted from  $\underline{A}$ .  $\underline{flavus}$  and chemically and physically characterized. (FC-106(C))

4. Microflora of Flue-Cured Tobacco and Their Effect on Quality. Growth of test fungi in inoculated cigarettes can be regulated by time, temperature, and the moisture content of the tobacco thus permitting cigarettes to be smoked on the smoking machine after progressive amounts of fungus growth.

The morphology of <u>Talaromyces</u> (<u>Penicillium</u>) <u>duponti</u>, a thermophilic fungus isolated from flue-cured tobacco, has been characterized.

Ten fungi, including six species of Aspergillus, were grown for 7 days at 25°C in continuous light on modified Czapek's agar in sealed desiccators in known mixtures of 02-C02 or 02-N2. Growth of all fungi increased significantly with each quantitative decrease of carbon dioxide or each increase in oxygen. The same fungi were grown in 02-N2 mixtures. At low 02 the fungi divided into two groups. A.niger, A. flavus, A. ochraceus, Alternaria and Penicillium were inhibited at 0% 02.

A. amstelodami, A. ruber, A. repens, C. herbarum, and Chaetomium were inhibited between 0.5 and 1% 02.

Bacteria present on tobacco leaves harvested in 1968 from three plant positions were studied. Lower leaves contained more bacteria than upper leaves. Within the confines of this test, yeasts constituted 25-30% of the organisms isolated. Staphylococcus epidermidis, a yellow-pigmented circular smooth translucent bacterial colony, was consistently isolated from tobacco samples. Based on cultural morphology, growth characters, and physiological tests, most of the yeasts are species of Cryptococcus, Candida, and Rhodotorula. (FC-110(A))

5. Microflora of Tobacco and Their Affect on Quality. The numbers and kinds of fungi isolated from 100 piles (total) of low and high stalk tobacco offered for sale in two warehouses within each of the five flue-cured tobacco belts indicated more Penicillium in tobacco from Kinston than from Rocky Mount, North Carolina, and in tobacco from Durham than from Warrenton, North Carolina. A. flavus was isolated more frequently from tobacco from Danville than from South Boston, Virginia. A. repens was isolated more frequently from low rather than high stalk tobacco in the Middle Belt.

In a replicated experiment, moisture contents of tobacco stored at 75, 80, 85, 87, and 95% relative humidity and 20 and  $30^{\circ}\text{C}$  were determined at weekly intervals for 4 weeks by four methods. Relative humidity, method and time were highly significant. The best of the four methods is 100 C/16 hours.

In a replicated experiment, tobacco inoculated with  $\underline{A}$ . repens was stored at 75, 80, 85, 87, and 95% relative humidity, at 20 and 30°C, and tested weekly for 4 weeks.  $\underline{A}$ . repens will not grow in tobacco for 4 weeks if stored at 20°C and at or below 80% relative humidity or 30°C at or below 75% relative humidity.

Fungi were identified from top and bottom leaves of field grown <u>Nicotiana rustica</u>, <u>N</u>. <u>sylvestris</u>, <u>N</u>. <u>glutinosa</u> and two varieties of <u>N</u>. tabacum. Eleven genera, including three species of <u>Aspergillus</u>,

were isolated, with <u>Alternaria</u> and <u>Penicillium</u> predominant. The number of fungi isolated from upper and lower leaves were 1757 vs. 1702, but some genera were more common on lower leaves. Host species and varieties were not different for the fungi isolated. (FC-126)

6. The Fate of Polyphenols in Tobacco During Air Curing. Methods for purification of polyphenol oxidases, the effect of virus infection and chemical agents on polyphenol oxidase and peroxidase activity, and the changes in polyphenols following virus infection are being investigated. A brown, high molecular weight pigment was found in young tobacco leaves. (FC-130(C))

## Publications - USDA and Cooperative Program

- RPA 702 PROTECT FOOD SUPPLIES FROM HARMFUL MICROORGANISMS
  AND NATURALLY OCCURRING TOXINS
- Fletcher, J. T., G. B. Lucas and R. E. Welty. 1967. Thermophilic fungi and bacteria isolated from tobacco. Phytopathology 57(5): 458-459 (Abstract). (FC-110(A))
- Welty, R. E. and G. B. Lucas. 1968. Fungi isolated from damaged flue-cured tobacco. Applied Microbiology 16:851-854. (FC-110(A))
- Welty, R. E., G. B. Lucas, J. T. Fletcher and H. Yang. 1968. Fungi isolated from tobacco leaves and brown spot lesions prior to and following flue-curing. Applied Microbiology 16:1309-1313. (FC-51(A))
- Welty, R. E. and G. B. Lucas. 1969. Fungi isolated from flue-cured tobacco at time of sale and after storage. Applied Microbiology 17: 360-365. (FC-110(A))

# PROGRESS REPORT OF THE HORTICULTURAL CROPS RESEARCH BRANCH MARKET QUALITY RESEARCH DIVISION AGRICULTURAL RESEARCH SERVICE

#### AREA 1

#### CITRUS AND SUBTROPICAL FRUITS

Research Activity Code: 1-12-404-14-403

# USDA and Cooperative Program

		0 - 1	- 27	. 77	77 77	1000
	•	Scientist	E Ma	an-Years	F.Y	1969
Location of Intramural Work	:_	Research	Pro	oblem Ar	ea:	
		404	:	501	:	Total
	:		:		:	
Citrus fruit other than limes	:		:		:	
California	:	3.0	:		:	3.0
Florida	:	3.8	:	1.0	:	4.8
Total Citrus	:	6.8	:	1.0	:	7.8
	:		:		:	
Avocados, mangos, limes and papay	as:		:		:	
Florida		0.2	:		:	0.2
	:		:		:	
Total	:	7.0	:	1.0	:	8.0
	:		: '		:	

Intramural program is supplemented by extramural support representing 0.3 SMYs at State Experiment Stations 1/2, and P.L. 480 funds in two countries totaling 129,000 U.S. dollars equivalent.

# Progress - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

## A. Quality Maintenance in Storage

1. CA storage of Florida citrus fruits. Valencia oranges stored for 3 months at 34° F. in air or in CA (15% oxygen plus 1% carbon dioxide) both in commercial storage and in laboratory storage had less than 3%

<sup>1/</sup> RPA 404 0.3

decay regardless of treatment. After an additional week at  $70^{\circ}$ , the commercially treated fruit had 20% decay and the laboratory treated fruit only 5%. Flavor of all fruit was good but CA-stored fruit were firmer and brighter.

Temple oranges after 5 weeks at 40° plus 1 week at 70°, had 15% decay in controls but only 2% decay in fungicide-treated lots. Both CA- and air-stored fruit had good to excellent flavor. Benlate, TBZ, and preand post-storage application of SOPP successfully reduced decay for 10 weeks. The flavor of CA lots was fair to good after 10 weeks but the distinctive Temple aroma was not retained.

The ethanol content of juice of all fruits increased during storage in direct relation to decreasing oxygen concentration in the storage atmosphere. Carbon dioxide, up to 5%, had no apparent effect but waxed fruit was consistently higher in ethanol than unwaxed fruit. Valencia oranges held under 100% carbon dioxide had higher ethanol in juice than those in 100% nitrogen. Temple oranges, held in 100% nitrogen for 2 weeks developed no off-flavor but after 3 weeks, off-flavor developed and ethanol content of juice had increased 170-fold. (HC-41)

2. CA storage of subtropical fruits. Avocados stored at  $50^{\circ}$  F. under a constant flow of 1.5-2.0% oxygen plus 10.0-10.5% carbon dioxide developed less decay than fruit stored in a static atmosphere of the same concentration or in air for 20, 30, and 40 days. Treatment with 1000 ppm Benlate dip resulted in only a slight reduction in anthracnose decay.

Decay was extensive in Keitt mangos stored in several controlled atmospheres at 55° F. A combination of 1000 ppm Benlate dip and hot water treatment reduced decay levels after 20 and 30 days of storage, but not to an acceptable level. The best controlled atmospheres tested were 5% oxygen plus 5% carbon dioxide and 10% oxygen plus 0% carbon dioxide.

Limes stored in 1% oxygen plus 99% nitrogen for up to 6 weeks at 50° F. maintained a thicker, darker green rind than fruit in air or in 10% oxygen plus 90% nitrogen. Storage in 1% oxygen reduced the amount of extractable juice. (HC-38)

3. Storage of Arizona lemons. Arizona lemons stored for 7 months at 57° F. under high humidity were in a marketable condition and sustained a weight loss of less than 5%. Decay was less than 1% in waxed fruit treated with 2,4-D. Yellowing was retarded by treating the fruit with 250 ppm gibberellic acid or by holding fruit in 10% oxygen. (HC-45)

#### B. Quality Maintenance during Transportation

1. Export shipment of California citrus fruits. The biphenyl residues in oranges were higher in non-vented cartons than in cartons with 1/2-and 1-inch vents. Aluminum foil barriers, placed on both sides of the biphenyl pad, decreased biphenyl residues in oranges more than barriers of wax paper or polykraft paper. Only small differences in biphenyl absorption by oranges resulted from addition and removal of biphenyl pads after various intervals during storage. Generally, reductions in biphenyl residues were accompanied by an increase in decay and soilage.

The amount of biphenyl absorbed by sound oranges was not affected by the amount of decayed fruit present during storage. Biphenyl vapor reduced sporulation and mycelial growth of Penicillium green mold markedly but absorbed biphenyl present in oranges did not.

Lemons shipped to Rotterdam, The Netherlands, in cargo containers were in excellent condition on arrival. Subsequent deterioration from decay and soilage was less than in the previous season shipments by the regular procedure. Biphenyl residues in lemons transported in containers were no higher than in regularly handled lemons. (HC-48)

## C. Postharvest Physiology

- 1. Ripening of mangos. This P.L. 480 research being conducted at the University of Baroda, India, has shown that extracts from unripe mangos inhibit the activity of most enzymes (amylase, invertase, cellulase, glucose-6-phosphate phosphatase, phosphofructokinase, and pectin esterase) during ripening of the fruit. The extracted ripening inhibitor present in unripe fruit is protein in nature and is inactivated by ethylene. Fruit starch hydrolysis to sugars during ripening was found to be less in the chilled fruit than in healthy fruit. Amylase activity in chilled tissue was less but pectin esterase and invertase activities were higher.

  (HC-63 A7-MQ-6)
- 2. Maturation and ripening of avocados. P.L. 480 research being conducted at the National and University Institute, Rehovot, Israel, showed that methanolic extracts of the mesocarp, seed coats, endosperm and seed of developing avocado fruit gave both growth promotive and growth inhibiting zones on chromatograms. Analysis with the barley endosperm bioassay indicated that gibberellin-like activity was very strong in the seed coat of vigorously growing fruit but had almost vanished at fruit maturity.

Cytokinin activity analyzed by the soybean callus tissue culture bioassay was very high in the endosperm and seed coat extracts but decreased with fruit development.

Ethylene had no noticeable effect on unharvested fruit but caused an acceleration of ripening of harvested fruit. (HC-65 - A10-MQ-2)

#### D. Postharvest Disease Control

- 1. Control of decay of Florida citrus fruits. Benlate was the most effective of six fungicidal treatments in 16 tests for the control of decay in freshly harvested oranges. An average of less than 1% decay developed in 3 weeks. Other treatments in descending order of decay control were: 5-aceto, 8 hydroxyquinoline sulfate; 2-aminobutane phosphate; thiabendazole (TBZ); hot water; and SOPP plus hexamine (standard treatment). Benlate was also most effective in tests with stored citrus fruit. TBZ, which was approved by the Food and Drug Administration in February 1969 for use on citrus, was more effective than SOPP in four commercial shipping tests. Benlate was effective in reducing postharvest decay when applied before harvest. Ethrel and ascorbic acid showed promise as agents for hastening on-tree color development. (HC-42)
- 2. Materials affecting germination and growth of decay organisms. Aldehydes of the C<sub>4</sub>-C<sub>9</sub> series were effective as fumigants for oranges inoculated with Penicillium spores. The shorter chain aldehydes caused some rind injury. The following molar concentrations of the aldehydes in culture media gave 100% inhibition of germination of Phomopsis citri spores: valeraldehyde, 0.017; hexanal, 0.0375; heptanal, 0.0009; octanel, 0.0019; and nonanal, 0.00051. All of these aldehydes except valeraldehyde have been reported isolated from orange tissue. (HC-44)
- 3. Effect of modified storage atmospheres on the growth of citrus decay organisms. A slight reduction of sporulation of Penicillium digitatum occurred when the carbon dioxide concentration was increased to 10%, whereas, an increase to 20% CO<sub>2</sub> caused a marked reduction. Little fungus growth occurred on fruit in 57% carbon dioxide but all fruit decayed normally when removed from the modified atmosphere to air. Valencia oranges which had been previously stored in 20% carbon dioxide had a slight off-flavor and those stored in 57% carbon dioxide were seriously off-flavor.

The taste of Valencia oranges stored in air at  $42^{\circ}$  F. was adversely affected when decaying oranges (inoculated with 5 common decay organisms) were present. The deterioration was greater at  $42^{\circ}$  in the presence of decayed fruit than at  $70^{\circ}$  in the absence of decayed fruit. (HC-46)

- 4. Decay related to mechanical harvesting. This study conducted under a cooperative agreement with the Florida Agricultural Experiment Station at Lake Alfred, Florida, showed that there was a significant increase in postharvest decay of fruit mechanically harvested with the "spindle harvester" and the "air shaker" harvester over that of hand-picked fruit. The increase in decay in the mechanically harvested fruit is attributed to damage of the abscission layer under the button. The increased decay was approximately doubled when the fruit was allowed to fall to the ground. Also, sunburn of "wind-rowed" fruit was a problem if the fruit remained on the ground for any length of time. A significant increase in decay occurred when the abscission chemical sodium cyclamate was used in conjunction with the "air shaker" harvester. Fumigation with 2-amino-butane (2 AB) vapor effectively controlled decay of harvested fruit.

  (HC-73A)
- 5. Antimicrobial action of biphenyl and derivatives. Research on this P.L. 480 project in Germany showed that biphenyl is slightly inhibitory to hexokinase, glycerol-1-phosphate dehydrogenase, and \(\beta\)-hydroxyacyl-CoA-dehydrogenase, and strongly inhibitory to phosphotransacetylase and glutumate dehydrogenase. Sodium orthophenylphenate (SOPP) has only a slight inhibitory effect on hexokinase and guanase but blocks phosphotransacetylase and glutumate dehydrogenase. The measurements indicating strong or blocking activities are assumed to be effective under the biological conditions of the use of the fungicide.

Naturally occurring orange and lemon oils were found to inhibit the metabolism of <u>Trichoderma</u> <u>lignorum</u> and when small quantities of SOPP (10-3 mol/1) were combined with 0.4% lemon oil, the respiration was completely blocked. The inhibiting effect of the oils increased strikingly with the addition of SOPP. (HC-67 - E10-AMS-3)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. Degreening Florida oranges and tangerines. Studies on Hamlin oranges indicated a slower initial rate of degreening at 70° F. than at 85°, but no difference was noted after 4 days of degreening. This indicates that temperatures below 85° might be useful during longer degreening periods early in the season. During a 1 or 2 day degreening period at 70°, the maximum degreening rate was reached with 1 to 5 ppm ethylene. During 4 days of degreening at 70° or 85°, the maximum response was obtained with 10 ppm ethylene. Responses to temperatures of 75° and 80° generally were intermediate between those at 70° and 85°.

Preharvest chemical treatments reduced the chlorophyll level in Robinson tangerines which might help reduce decay and other problems resulting from postharvest degreening of fruit of this variety. (HC-43)

#### Publications - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### Quality Maintenance in Storage

- Craft, C. C., G. Duncan, and D. Fouse. 1968. Respiratory activity of small lemons and lemon tissue as influenced by modified atmospheres. Proc. Amer. Soc. Hort. Sci. 93: 173-185. (HC-45)
- Hatton, T. T., Jr., and W. F. Reeder. 1968. Quality of Persian limes after different packinghouse treatments and storage in various controlled atmospheres. Proc. Tropical Region, Amer. Soc. Hort. Sci. 11: 23-32. (HC-38)
- Norman, Shirley, and C. C. Craft. 1968. Effect of ethylene on production of volatiles by lemons. HortScience 3(2): 66-69. (HC-47)

#### Quality Maintenance during Transportation

Norman, Shirley, G. L. Rygg, and D. C. Fouse. 1968. Biphenyl residues in oranges: Effect of biphenyl dosage and pad placement. ARS 51-26. (HC-48)

#### Postharvest Physiology

- Cooper, W. C., G. K. Rasmussen, and J. J. Smoot. 1968. Induction of degreening of tangerines by preharvest applications of ascorbic acid and other ethylene-releasing agents. The Citrus Industry 49(10): 25-27. (HC-42)
- Hatton, T. T., and W. F. Reeder. 1969. Stylar-end breakdown in Persian limes as influenced by temperature and bruising. Proc. Fla. State Hort. Soc. 81: 344-349. (HC-39)
- Mattoo, A. K., V. V. Modi, and V. V. R. Rehdy. 1968. Oxidation and carotenogenesis regulating factors in mangos. Indian Jour. Biochem. 5(3): 111-114. (HC-63 A7-MQ-6)
- Mattoo, A. K., and V. V. Modi. 1969. Ethylene and ripening of mangos. Plant Physiology 44(2): 308. (HC-63 A7-MQ-6)

#### Postharvest Disease Control

Davis, Paul L. 1968. Determination of solubilities of  $C_5$ - $C_9$  aldehydes in water by gas chromatography. J. Gas Chromatography 6: 518-519. (HC-44)

- Rehm, H. J., P. Wallnofer, and G. Herbig. 1968. Zur Kenntnis der antimikrobiellen wirkung von biphenyl and derivaten des biphenyl. IV. Mitteilung, Wirkung auf den Kohlenhydratstoffwechsel von Pilzen. A. Lebensmittel-Unters. u. Forsch. 136: 167-175. (HC-67 E10-AMS-3)
- Rehm, H. J., and J. Baltes. 1968. Zur Kenntis der antimidrobiellen Wirkung von biphenyl und derivaten des biphenyls. VI. Mitteilung, Wirkung auf Enzyme des Kohlenhydratstoffwechsels. A. Lebensmittel Unters. u. Forsch. 137: 295-303. (HC-67 E10-AMS-3)
- Srivastava, M. P. 1968. Utilization of lactose and formation of synthetic oligosaccharide by isolates of <u>Botryodiplodia theobromae</u>. Proc. Natl. Acad. Sci., India, 38(B), I. (HC-63 A7-MQ-6)
- Wallnofer, P., and H. J. Rehm. 1968. Zur Kenntnis der antimikrobiellen wirkung von biphenyl und derivaten des biphenyls. III. Mitteilung, Einfluss von biphenyl und Na-o-phenylphenolat auf die Entwicklung Citrus--verderbender Schimmelpilze. Zentralbl. f. Bakt. II, 122: 25-41. (HC-67 E10-AMS-3)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

#### Objective Measurement of Quality

Jahn, Otto L., W. G. Chace, Jr., and Randall H. Cubbedge. 1969.

Degreening of citrus fruits in response to varying levels of oxygen and ethylene. J. Amer. Soc. Hort. Sci. 94(2): 123-125. (HC-43)

AREA 3

DECIDUOUS FRUITS AND TREE NUTS

Research Activity Code: 1-38-404-14-402

#### USDA and Cooperative Program

Scientist Research				1969
	Probl	A		
. /.0/.		em Area	_:	
<b>:</b> 404	:	501	:	Total
•	:		:	
•	:		:	
2.0	:	1.0	:	3.0
1.0	:	0	:	1.0
3.0	:	1.2	:	4.2
1.0	:	0	:	1.0
2.0	:	0	:	2.0
9.0	:	2.2	:	11.2
	:		:	
•	:		:	
1.0	:	0	•	1.0
0.5	•	0	•	0.5
0.2	:	0	:	0.2
1.7	:	0	:	1.7
10.7	:	2.2	:	12.9
	2.0 1.0 3.0 1.0 2.0 9.0	2.0 1.0 3.0 1.0 2.0 9.0 1.0 0.5 0.2	2.0 1.0 1.0 0 3.0 1.2 1.0 0 2.0 0 9.0 2.2  1.0 0 0.5 0 0.2 0 1.7 0	2.0 1.0 1.0 1.0 1.0 1.0 0 1.2 1.0 0 0 1.2 1.0 0 0 1.2 1.0 0 1.2 1.0 0 1.2 1.0 0 1.2 1.0 0 1.2 1.0 0 1.2 1.0 1.0 1.0 1.0 1.0 1.0 1.0 1.0 1.0 1.0

Intramural program is supplemented by extramural support representing (a) 0.8 SMYs at State Agricultural Experiment Stations  $\frac{1}{2}$ , (b) 0.5 SMYs at other U.S. institutions  $\frac{2}{2}$ .

#### Progress - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### A. Quality Maintenance in Storage

1. CA storage of stone fruits. Peaches stored 6 weeks in a controlled atmosphere (CA) of 1% oxygen, 5% carbon dioxide, and 94% nitrogen (1-5 atmosphere) at 32° F. ripened with a better internal appearance than fruit stored in air.

<sup>1/</sup> RPA 404 0.8

<sup>2/</sup> RPA 404 0.5

Prestorage dips in Benlate fungicide reduced decay in both CA and air storage, but caused a skin browning of the Redhaven and Redskin varieties. A prestorage hot water dip (125° F. for 2½ minutes) also caused a skin browning of the Redhaven variety after 6 weeks' storage.

Less mature Redglobe peaches (11 to 15 lbs. pressure at harvest) retained better quality in air and CA storage than more mature (9 lbs.) fruit. Fruit from the CA storage after 6 and 9 weeks had slightly better internal appearance and less decay than fruit stored in air.

Brown rot inoculated peaches treated with Benlate fungicide or hot water before storage in CA or air at 32° F. developed less decay during 6 weeks' storage and subsequent ripening at 65° than non-treated fruits. However, only Benlate was effective after 9 weeks' storage.

Brown rot inoculated nectarines reacted the same as peaches when treated with Benlate and hot water before air or CA storage. However, hot water treatment was effective in controlling brown rot of nectarines stored for 9 weeks in 1% oxygen with 5% carbon dioxide.

Late Le Grand nectarines stored in 1-5 controlled atmosphere at 32° F. for 9 weeks ripened with a better internal appearance than fruit stored in air or in air with 5% carbon dioxide. Hot water dips caused moderate to severe skin browning when fruit was held in controlled atmospheres for more than 6 weeks. Benlate-dipped nectarines had reduced decay after 9 weeks in all atmospheres with no injury to the skin. However, fruit stored 12 and 15 weeks developed some skin browning and poor internal appearance in all atmospheres. (HC-3)

- 2. CA storage of strawberries. Carbon dioxide atmospheres of 21-23% in strawberry pallets during air shipment reduced decay from 4.1% in control lots to 1.9% one day after arrival in New York and from 22.7% to 9.4% two days after arrival. Laboratory tests showed that Rhizopus rot was significantly reduced by carbon dioxide concentrations above 10% and gray mold rot was significantly reduced at carbon dioxide levels above 20%. No off-flavors developed in carbon dioxide levels below 30%. Off-flavors occurring in carbon dioxide above 30% disappeared when the berries were placed in air for 24 hours. (HC-27)
- 3. Storage of Chinese gooseberries. The flesh color of Chinese gooseberries did not change during storage at 32° F. for 0, 4, or 8 weeks. Fruit that was firm initially changed more in flesh color than fruit that was soft at harvest. The more mature (soft) fruit had slightly higher soluble solids and pH than the less mature (firm) fruit at harvest and this did not change appreciably during storage or ripening.

The firmness of fruit decreased as storage and ripening time increased. Weight-loss was negligible during storage in polyethylene bags, but averaged 2.0% after 1 week and 3.5% after 2 weeks ripening at  $70^{\circ}$ . Decay was negligible in all lots. (HC-68)

- 4. Storage of new pear varieties. There was no apparent differences in pH between the 1967 and 1968 stored fruit although the soluble solids and total acidity of the fruit tended to be higher in 1968. Pressure tests indicated a lesser degree of ripening in storage than occurred in 1967. (HC-58A)
- 5. Cranberry storage. About two-thirds as much spoilage developed in Massachusetts cranberries held in 38° storage with a high relative humidity as was found in berries held in an air-cooled storage averaging 50° F. and 63% relative humidity. Cranberries stored at 33° but interrupted each month by 1 day at 70° had about two-thirds as much spoilage as those held in either continuous 33° storage or in air-cooled storage. Decay was slightly greater in berries from interrupted storage than from continuous storage but physiological breakdown was less from the interrupted storage. (Exploratory)

#### B. Quality Maintenance during Transportation

1. Air shipment of California strawberries. Curtain-coated fiberboard pallet covers with taped seams, or with folded, strapped closures effectively maintained high carbon dioxide atmospheres in strawberry pallets during air shipment.

Strawberries packed in plastic-mesh baskets averaged 9.6% cutting damage when shipped from California to eastern markets as compared to 3.5% cutting when packed in paper-pulp baskets. Overall damage (cutting plus bruising) on arrival averaged 18.2% in plastic-mesh and 13.6% in paper-pulp baskets. (HC-27)

2. <u>Precooling strawberries</u>. Forced-air precooling reduced the half-cooling time for palletized loads of berries (in fiberboard trays with plastic baskets) from the average 2 hours required in an ordinary precooling room to only 30 minutes.

When strawberries were packed in plastic-mesh trays they cooled almost twice as fast in an ordinary room precooler as did berries packed in fiberboard trays. Also, berries in plastic baskets cooled approximately 20% faster in an ordinary room precooler than those berries packed in pulp baskets. However, cooling rates for fruit in both types of baskets were similar when cooled in a forced-air precooler. (HC-69)

#### C. Postharvest Physiology

- 1. Strawberry ripening as affected by pressure. Subjective inspections of three strawberry varieties indicated that the ripening of strawberries was progressively delayed with pressure decreases at 35° F. in hypobaric chambers. (HC-9)
- 2. Scald control for western apples and pears. Ethoxyquin wraps, when applied immediately after harvest and after delays of 2, 4, 6 and 8 weeks, gave complete control of scald of Golden Delicious apples stored 7 months at 32° F. Ethoxyquin when applied as a dip or wrap, or mixed with captan in a dip, controlled scald on Anjou pears. Captan alone increased scald. Unlike previous results, hot water failed to control scald. However, hot water was more effective in reducing stem decay than the fungicides that were tested.

After 6 months cold storage, scald of Starking Delicious apples was significantly reduced in lots of fruit hot water (125° F.)-treated 1 and 4 weeks after harvest. However, scald-reduction in fruit treated 4 weeks after harvest was not significant.

Heat injury occurred on apples treated at a hot water temperature of 125° 8 weeks after harvest and at 130° or 135° at one, four or eight weeks after harvest. In general, susceptibility to heat injury increased as the period between harvest and treatment was extended.

(HC-54)

3. Effect of preharvest environment on postharvest quality of apples. Storage scald appeared to be correlated with the chlorophyll content of the apple fruit. Two types of scald were found on Golden Delicious apples--one related to maturity, and the other related to the initiation of senescence.

Soluble solid content was not influenced and only a small increase in acidity and firmness occurred after 6 months' storage of apples sprayed in the field with 1000 ppm Alar.

Ethrel, applied a few days before harvest, did not affect chlorophyll loss, firmness, soluble solids, or titratable acidity of fruit examined 3 and 8 months after harvest. (HC-55)

4. Effect of preharvest environment on postharvest quality of pears. The abnormally low temperatures occurring in Oregon and California just prior to the 1968 harvest, which caused severe premature ripening and fruit droppage of pears, substantiated the 1967 and 1968 results of tests in the Hood River Valley of Oregon. In these tests, premature

ripening and dropping of fruit occurred when limbs were caged and artificially cooled, but no droppage occurred when caged limbs were artificially heated. Fruit in heated cages were approximately 50% larger in size than fruit in cooled cages. The respiratory activity was much higher in fruit from cooled than from heated enclosures. Data indicate that changes in respiratory activity can be detected after relatively short periods of exposure to unfavorable temperatures. Also, temperature data indicate that short exposures to critically low temperatures may be as effective in initiating ripening as larger exposures to deficiency in accumulated heat units.

(HC-57A)

5. Scald and oxidative browning of red tart cherries. Aeration of red tart cherry soak water does not alleviate scald at 50-70° and causes increased oxidative browning. Delaying harvest until cherries are fairly mature decreases their susceptibility to scald and oxidative browning. (Exploratory)

#### D. Postharvest Disease Control

- 1. Blue mold rot of apples. When applied within 5 hours after wounding and inoculating with the causal fungus, heated (45 seconds at 130° F.) 500 ppm solutions of Benomyl (Benlate) and Thiabendazole (TBZ) controlled blue mold rot in both punctured and bruised Jonathan, Golden Delicious, and Red Delicious apples. However, unheated solutions of the same solutions controlled the rot only in punctured apples. When applied 24 hours after wounding and inoculating, the heated chemical solutions controlled rot in only bruised apples. Hot water (45 seconds at 130° F.) alone controlled rot only in bruised apples given the delayed treatment. Heated water and heated Benomyl and Thiabendazole solutions sometimes caused a scald-like skin injury to develop on apples during 3 months of storage at 32° F. Apples treated with heated Benomyl or TBZ solutions had a higher incidence of internal breakdown than fruit treated with comparable unheated solutions. (HC-2)
- 2. Decay control for strawberries. Strawberries treated with hot air (108° for 20 minutes), or dipped in dehydroacetic acid (DCNA) or captan before holding for 36 hours at 59° were not significantly different from the untreated berries at first, but after an additional 24 hours at 59°, there was significantly less decay than in the untreated berries. Still later, the berries originally held at 35° had significantly less spoilage than those treated with captan and held at 59°.

Combined losses from decay caused by species of <u>Rhizopus</u>, <u>Botrytis</u>, <u>Alternaria</u>, <u>Cladosporium</u>, and <u>Phytophthora</u> were significantly less in berries held for 36 hours at 35° F. in a normal atmosphere or at 59° in 20% carbon dioxide atmosphere than in check lot of berries held at 59° in a normal atmosphere.

Decay was not controlled on strawberries held for 4 days at 65° F. after they had been dipped in solutions of 100 or 500 ppm of Benlate, Thiabendazole (TBZ), Botran or Penn Salt chemicals (TD 5056, TD 1534, and TD 1604). The Penn Salt chemicals turned the green calyx of the berries a gray-brown color and caused some bleaching of the fruit.

(HC-28)

- 3. Effect of chemicals and heat on growth of certain fungi. Rhizopus stolonifer and Monilinia fructicola cultured on a glucose-yeast extract medium containing 2,6-dichloro-4-nitroaniline (DCNA) produced variants which grew well in the presence of 100-1000 ppm DCNA. Growth of the parent cultures was reduced by 1 ppm DCNA and completely inhibited by 10 ppm DCNA. Spores of both parent and variant cultures were equally sensitive to hot water treatments. (HC-30)
- 4. Effect of mechanical harvesting on blueberry postharvest decay. Although fruit obtained with the mechanical shaker showed a greater range in ripeness and size, there was no obvious difference in decay development when compared with hand-harvested fruit. (HC-51)
- 5. Control of postharvest rot of stone fruits. Benomyl (Benlate) and Geigy 20092 solutions controlled brown rot on inoculated peaches about as effectively as treatment in 125° F. water for  $2\frac{1}{2}$  minutes. Both chemicals controlled decay development at concentrations of 0.125, 0.5, and 1.0 percents, but neither chemical controlled rhizopus decay. However, when used in 125° water, both chemicals controlled brown rot and rhizopus decay, but neither solution was consistently better than 125° water alone.

Reports that peaches with captan residues are injured when treated with hot water were confirmed. Fruit dipped in captan concentrations of 1, 2, and 4 pounds active per 100 gallons of solution then treated for 10 minutes in 122° F. water or for 2½ minutes in 125° F. water developed tan streaks in the skin giving a mottled surface appearance. Injury to hot water-dipped peaches from trees sprayed with 2 pounds/100 gallons captan showed most severe injury when the fruit was picked and dipped 2 hours after fungicide application, only slight dulling occurred when dipped 24 hours after application, and no injury was observed when dipped 48 hours after the captan spray was applied. The use of a supplemental wetting agent in the captan dips intensified the injury. (HC-4)

Combined hot water and 2,6-dichloro-4-nitroaniline (DCNA) treatments of peaches, plums, and nectarines were more effective in controlling post-harvest decays than either hot water or DCNA alone. When 225 or 450 ppm DCNA was added to the hot water, equivalent inhibition of lesion dévelopment on fruit inoculated with Monilinia or Rhizopus was possible with

treatments of half the exposure times necessary with hot water alone. Naturally occurring decay was reduced from a mean of 65 percent in untreated peaches and nectarines to 7.8 percent in lots immersed for 1.5 minutes in 125° F. water with 225 ppm DCNA. The residues of DCNA on the fruit after the treatment were equivalent to residues from treatments with 900 ppm DCNA in unheated water. (HC-75)

6. Role of enzymes and microorganisms in death of vegetable tissues. Ethyl acetate extracts of leaves or stems of Bartlett pear trees inhibited the growth of Erwinia carotovora more than the growth of E. amylovora. Heavy suspensions of E. carotovora injected into the soft stem tissue of budded Bartlett pear trees in the greenhouse caused a blackening of the injected tissue similar to fire blight. However, the affected area did not enlarge and the tissue did not ooze fluid as is common with fire blight. The black tissue reaction was not produced in pear trees injected with washed cells of  $\underline{E}$ . carotovora.

Pectin lyase and cellulase enzyme activity was detected in blue mold-rotted tissue of Red Delicious apples, but no phosphatidase activity was detected in the rotted tissue. (HC-10)

7. Identification of marketing diseases. Kiwi fruit (Actinidia chinensis--Isolations from decayed ripe fruit in the Chicago market showed the presence of bacteria, yeasts, and the fungi Penicillium, Phoma, Phomopsis, and Gloeosporium. Infection, presumably occurring prior to shipping, displayed external symptoms of dark spots, mold growth, and exudation on the fruit surface. Isolations have demonstrated the pathogenicity of Phomopsis.

Peaches--Bruises and mechanical injuries were found to be responsible for as high as 7½ percent of the wholesale and retail waste in peaches. Isolations showed that many of the bruised and injured areas were rotted by Geotrichum candidum indicating that peaches may be contaminated during handling operations and do not decay until almost ripe. Apparently bruised peaches contaminated with Geotrichum have a shelf life of from one to several days less than peaches similarly bruised but free of contamination. (HC-23)

8. Losses in fruits during marketing. Apples-Red Delicious apples from the Northwest averaged about 5% loss through the consumer level in the Chicago market and about 4% in the New York market. Bruise damage accounted for most of the loss in the Chicago market and at the retail level in the New York market. Decay caused most loss at the wholesale level, and internal breakdown at the consumer level in the New York market. Appalachian Red Delicious apples averaged about 2% market loss in the New York metropolitan market mostly due to bruising.

<u>Peaches--Losses</u> through the consumer level averaged more than 35% and 15% respectively, in the Chicago and New York markets. In both markets, physical bruising was the principal cause of loss at the wholesale and retail levels, whereas brown rot caused most loss at the consumer level.

Strawberries --Market losses for California strawberries over an 8-month marketing period in the Chicago market averaged over 47%. Physical injury was the principal cause of losses at all levels with greatest loss occurring at the consumer level. Losses in the New York market, which averaged over 31%, were mostly caused by gray mold rot. However, bruising and overmaturity caused over 7% of the 20% loss occurring at the consumer level. (HC-19 and 24)

9. Storage sanitation. Populations of microorganisms in storage rooms, controlled atmosphere chambers, and storage containers were greatly reduced by washing the surfaces with soap and water. The populations are generally reduced to near zero when rooms or containers are further disinfected with 70 percent alcohol. (Exploratory)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. Eastern apples. Freshly harvested Red Delicious apple sports (Bisbee and Red Prince) were sorted into 6 categories according to their stiffness coefficients, f<sup>2</sup>m, derived from sonic resonance frequencies and mass of the apple. After 16 weeks of storage at 32° F. no significance was found between stiffness coefficients and the following measurements made on an Instron Universal Testing Machine using (1) Magness-Taylor probe with apple skin removed, (2) 1-inch sphere with apple skin removed and (3) 1-inch sphere with skin intact. No significance between stiffness coefficients and taste panel ratings for general texture, firmness, or toughness was found. Correlations between measurements made with the Magness-Taylor probe and the following taste panel ratings were much higher: general texture (r=0.879); firmness (r=0.863; toughness (r=0.848). Correlations of measurements made with the 1-inch spheres, skin intact or removed, with panel ratings were considerably lower than those for the Magness-Taylor measurements. (HC-11)
- 2. Western apples. The chlorophyll content of Northwest Red and Golden Delicious apples and their external fruit color were found to be closely related to the nitrogen level in the tree. Also, the dessert quality after storage was judged to be inversely related to the relative chlorophyll content of the fruit. Inversely related to the apple chlorophyll content were: (1) the acid content of fruit harvested 135 and 145 days after bloom, (2) soluble solids, and (3) the flesh firmness of fruit harvested 135 and 145 days after bloom. (HC-53)

#### Publications - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### Quality Maintenance in Storage

- Anderson, R. E., C. S. Parsons, and W. L. Smith, Jr. 1967. CA storage of peaches. Proc. Natl. Peach Council, 26th Ann. Convention and Trade Show: 118-123. (HC-3)
- Harris, C. M., F. M. Porter, and J. M. Harvey. 1969. Market quality and precooling rates of strawberries packed in various containers. USDA, MRR 851. (HC-69)
- Kushman, L. J. and W. E. Ballinger. 1968. Commercial forced-air cooling of blueberries in North Carolina. Proc. Amer. Soc. Hort. Sci. 92: 788-792. (HC-51)
- Lutz, J. M. and R. E. Hardenburg. 1968. The commercial storage of fruits, vegetables and florist and nursery stocks. USDA, Agr. Handb. 66, 94 pp. (HC-1)
- Smith, W. L., Jr., R. E. Anderson and C. S. Parsons. 1969. Controlled atmosphere storage and decay of peaches and nectarines. (Abs.) Phytopathology 59: (HC-3)
- Worthington, J. T., R. E. Hardenburg and H. C. Vaught. 1969. A comparison of the keeping quality of Jonathan and of Red Delicious apples stored in field boxes or prepackaged in film bags. USDA, ARS 51-27, 16 pp.

#### Quality Maintenance during Transportation

- Harvey, John M. 1969. Strawberries to Europe by air: Special packaging and containers help maintain quality. United Fresh Fruit & Vegetable Assn. Yearbook, pp. 32-36. (HC-27)
- Harvey, John M. 1969. Postharvest handling and transportation reviewed. The Packer 76(18): 6B.

#### Postharvest Physiology

Kushman, L. J. and W. E. Ballinger. 1968. Acid and sugar changes during ripening of Wolcott blueberries. Proc. Amer. Soc. Hort. Sci. 92: 290-295. (HC-51)

#### Postharvest Disease Control

- Burton, C. L. and W. R. Wright. 1969. Sour rot disease of peaches on the market. Plant Disease Reporter 53: 580-582. (HC-23)
- Ceponis, M. J., J. Kaufman and J. E. Butterfield. 1969. Moldy carpels in Delicious apples on the greater New York market. Plant Disease Reporter 53(2): 136-138. (HC-18)
- Smith, W. L., Jr. 1969. Decay control of peaches. The Maryland Fruit Grower 39(2): 41-44. (HC-4)
- Spalding, D. H. and B. C. Smale. 1969. Comparative interactions of Erwinia carotovora and Erwinia amylovora with pear and potato tissues. Plant Disease Reporter 53: 255-256. (HC-10)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

Ballinger, W. E., J. N. Yeatman and L. J. Kushman. 1968. Progress in maturity measurement of blueberries using light transmittance techniques. HortScience 3(2): 99.

AREA 9

#### POTATOES

Research Activity Code: 1-11-404-14-404

#### USDA and Cooperative Program

Location of Intramural Work	:	Scientist Man Years, FY 1969
Minnesota	•	1.0
Maine		1.0
Maryland (Beltsville)	•	0.2
California		0.2
Total	:	2.4

#### Progress - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### A. Quality Maintenance in Handling and Packaging

1. <u>Tuber bruising</u>. In Maine, Kennebec potatoes exhibited no loss of tuber quality when subjected to a water flume-stone trap before storage in pallet boxes or bulk bins held at 45° F. Although the Russet Burbank variety did not lose quality when flumed and stored in pallet boxes, 25.3% Fusarium tuber rot developed when tubers were flumed and placed in 45° bulk storage. However, those tubers treated with a 2-minute dip of DAC 2787 and M-45 solutions after fluming had only 7.1 and 12.8% Fusarium rot, respectively.

A stationary air-vacuum stone separation machine located at the storage was satisfactory for stone removal for 3 of the 4 stone levels tested. Excessive tuber bruising resulted for bulk loads of potatoes containing over 4.2% stones by weight for a 120 cwt. bulk potato body.

Tubers flumed in metal flumes showed an increased amount of bruising as the angle between the supply flume and the main flume was decreased. Thirty-two and a half percent of the tubers were bruised at an angle of 90°; whereas, at a 60° angle, bruised tubers were increased to 37.6% and at a 45° angle, 41.4% of the tubers were bruised. Warming the tubers to 65° F. before fluming at the 45° angle decreased bruised tubers to 8%. Flume water temperature had no effect on the amount of bruising.

Tuber maturity and bruise resistance were found to be correlated. Tubers allowed to mature 3 weeks in the field before harvest had a bruise resistance index of 10.9-11.5, whereas, immature tubers showed an index of 7.5-9.5. Tuber maturity had no effect on weight loss, pressure bruising, or internal black spot. (HC-50)

#### B. Quality Maintenance in Storage

1. Storage of precut seed. Seed potatoes of two varieties, Russet Burbank and Kennebec, washed before cutting and cut monthly from December to May, had a lower percentage of Fusarium seed piece decay than non-washed potatoes. Although the rate of plant emergence was greater for the February through April cut seed than for seed cut at planting (due mainly to greater sprout growth at planting), the final stands for all treatments were approximately equal.

In other tests, the seed of both varieties remained in excellent condition for 2 months after precutting when treated with Polyram or M-45 dust and stored in bulk bins until planted. However, tubers of the Kennebec variety from the bulk bin storage had poorer emergence and stand than fresh cut seed. Precut Russet Burbank seed which had not been treated with a fungicide dust before storage had a high incidence of seed piece rot resulting in a poor stand, 61% compared to 98% for treated seed. (HC-49)

2. <u>Warming potatoes in storage</u>. When the temperature of bulk stored potatoes was increased in a closed cross-flow ventilation system without addition of moisture, the relative humidity dropped to a point where desiccation of tubers and pressure bruising was severe. (HC-50)

# C. Quality Maintenance during Transportation

1. <u>Disorders in transit</u>. Potatoes shipped from California to New Jersey averaged 70° F. tuber temperature at time of loading and 46° during the transit period. Internal blackspot was found to be the most prevalent disorder at destination with mechanical damage (cuts, splits, and bruises) the second most prevalent problem encountered. Soft rot was of minor importance in the test shipments. (HC-33)

2. <u>Bulk shipment</u>. Results from 17 test car shipments from the Red River Valley showed that the 1200 hundredweight converted Mechanical Refrigerator car, and the 1800 hundredweight center flow Cushionaire car were generally superior to the 600 hundredweight R.S. Reefer in both reduction of bruising and disease development in transit. Chip quality of processing potatoes was also improved over that in the R.S. Reefer. (HC-25)

#### D. Postharvest Disease Control

1. Losses during marketing. Mechanical damage and decay were the leading causes of losses in Maine Katahdin potatoes on the New York market. A 1.3% average loss was found at the wholesale level and 3.6% at the consumer level.

Fusarium tuber rot, bacterial soft rot, and mechanical bruising caused most of the losses to California prepackaged Long White potatoes on the New York market. Losses averaged 0.5% at the wholesale level and 4.6% at the consumer level. On the Chicago market, California Long White potatoes had an average loss of 2.31% at wholesale and 3.55% at the consumer level. Most losses were from physical damage.

A 7-month survey of Red River Valley red potatoes on the Chicago market showed an average loss of 4.87% at wholesale, 0.42% at retail, and 3.01% at the consumer level. The losses were mostly from physical damage.

(HC-19 and 24)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

l. Measurement of chip color. A new, simple and inexpensive procedure was developed to prepare flat potato chips directly from raw stock for color evaluation in inspection. Panelists' ranking of chips from lightest to darkest and the order of the Hunter L values from highest to lowest were identical. Color measurement of flat chips shows a significant increase in Hunter  $b_L$  values when UV light was included in the measuring beam. At the same time, L and  $a_L$  values decreased slightly. Mean panel scores were similar when judgments were made under light fixtures designed to include or exclude UV energy.

Correlation coefficients of color parameters and panel scores were similar whether samples were corrected for light scattering or not. Better visual match between potato chip and Munsell colors was obtained after correction of spectral data for light scattering. (HC-14)

#### Publications - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### Quality Maintenance in Storage

- Marlowe, G. A., Jr., D. N. Wright, J. C. Bishop, V. H. Schweers, et al. 1969. Preliminary Report: Potato sequence analysis, 1968. Potato Growers Assn. of Calif., Inc. Yearbook, pp. 115, 117, 119. (HC-33)
- Stewart, J. K. 1969. Preliminary Report: Potato sequence analysis study U.S. Department of Agriculture participation. Potato Growers Assn. of Calif., Inc. Yearbook, pp. 119, 121. (HC-33)

#### Postharvest Physiology

Stewart, J. K. 1969. Temperature effects on chip color of Kennebec potatoes. Potato Growers Assn. of Calif., Inc. Yearbook, pp. 87, 89, 91. (HC-33)

#### Postharvest Disease Control

Spalding, D. H. and B. C. Smale. 1969. Comparative interactions of Erwinia carotovora and Erwinia amylovora with pear and potato tissues. Plant Disease Reporter 53: 255-256. (HC-10)

#### AREA 11b

#### CUT FLOWERS AND ORNAMENTALS

Research Activity Code: 1-25-906-14-407

#### USDA and Cooperative Program

Location of Intramural Work	:	Scientist Man Years, FY 1969			
Florida	:	1.0			
California	:	0.5			
Maryland (Beltsville)	: :	1.0			
Total	:	2.5			

#### Progress - USDA and Cooperative Program

RPA 906 - CULTURE AND PROTECTION OF ORNAMENTALS AND TURF

#### A. Quality Maintenance in Storage

- 1. <u>Carnations</u>. Carnations, cut as 3/4- to 1-inch buds were shipped from California and Colorado to Maryland then held at 75° F. in preservatives. Loose buds opened in 1-2 days after arrival, whereas, tighter buds required 3-4 days. Buds which were open upon arrival had a mean vase life of 13-15 days when held in a preservative at 70° F.; 12-14 days after storage for 1 week at 40° F. before opening; and 7-14 days when stored 3 weeks at 32-33° F. Some lots were injured after 3 weeks at 32-33° and never opened well. "Cornell solution" usually produced the largest blooms with the longest life, but Everbloom (2%) and a solution containing 3% sucrose, 400 ppm 8-hydroxyquinoline citrate and 300 ppm Alar were also satisfactory preservatives. (Cooperative with T&FRD.)
- 2. <u>Iris</u>. 'Wedgewood' iris bulbs stored 46 days at  $45^{\circ}$  F. in 100% nitrogen or in 99.75% nitrogen plus 0.25% oxygen exhibited no external injury but produced flowering plants  $2\frac{1}{2}$  to 3 weeks later than bulbs stored in air. Plants from bulbs stored in high-nitrogen were slightly taller, excessively leafy, and weighed 35% more than plants from airstored bulbs.

"Blue ribbon" bulbs lost 6.4% weight during 7 weeks at 50° F. in air. Most bulbs were sprouted and rooted and 25% were infected with Penicillium. Bulbs held the same period in a controlled atmosphere of 100% nitrogen or in 99.75% nitrogen plus 0.25% oxygen, lost only 4.4% in weight and were in good condition. Bulbs stored in high nitrogen germinated well but much later than those stored in air. (Cooperative with CRD.)

- 3. Easter lilies. Lily bulbs stored well for 8 weeks at 40° F. but those stored in 100% nitrogen or in 99.75% nitrogen plus 0.25% oxygen produced shorter plants and bloomed 2-3 weeks later than those stored in air. Plants from bulbs stored in CA also had 47% more leaves and 59% more blooms than plants from bulbs stored in air. (HC-8)
- 4. Roses. Vase life of "Better Times" roses was prolonged by holding stems in water at pH 3-4. Low pH prevented vascular blockage and allowed greater uptake of water. Eight-hydroxyquinoline citrate (8-HQC) was more effective at pH 4.0 than at 6.0 but sucrose prolonged vase life at any pH. (HC-71)
- 5. <u>Gladiolus</u>. Vase life of gladiolus spikes was slightly benefited when held in 8-HQC plus sucrose for 2-4 hours prior to transfer to water. Spikes pre-treated in 8-HQC and sucrose wilted less after 4 days than spikes held in water. Gladiolus spikes held 24 hours at 110° F. in 8-HQC-sucrose solution were more resistant to wilting than spikes held in 70° solutions. (HC-71)
- 6. <u>Chrysanthemums</u>. Chrysanthemums harvested as buds and allowed to open in 8-HQC plus sucrose had a carbohydrate content similar to flowers opened intact on the plant. Both harvested and intact buds increased in carbohydrate and enlarged at equal rates. Buds held in water did not enlarge or increase in carbohydrate content.

The commercial flower preservative, "Everbloom," was as effective as 8-HQC-sucrose for opening freshly harvested chrysanthemum flower buds, but "Roselife" and "F.M." preservatives were not as effective.

The sucrose fraction of 8-HQC-sucrose was responsible for foliar chlorosis in chrysanthemums. This condition was controlled by holding stems in solutions containing low concentrations of sucrose. 8-HQC caused discoloration of the basal portion of chrysanthemum stems but discoloration did not appreciably shorten vase life. (HC-71)

# B. Quality Maintenance in Transit

Chrysanthemum flower buds shipped from Bradenton, Florida, to Washington, D.C., through commercial channels were successfully open at destination. These tests substantiated earlier laboratory results. (HC-71)

#### C. Postharvest Physiology

1. <u>Color retention in cut flowers</u>. The Hunter Color--Color Difference Meter was found to be a more effective instrument for measuring rose petal color than a Beckman reflectometer.

The intensity of red petal color of freshly harvested roses was reduced when stems were held in sucrose for several days. Color reduction was not true "fading" but was due to increased petal size (area) with no appreciable loss of anthocyanin. (HC-70)

- 2. Chrysanthemums. Flower buds allowed to open in 8-hydroxyquinoline citrate (8-HQC) plus sucrose or held on the plant had similar amounts of anthocyanin in the petals. However, buds held on the plant were more intensely colored. Those buds held in 8-HQC-sucrose had greater petal area than buds held on the plant thus producing a less intense color. (HC-70)
- 3. <u>Carnation response to ethylene</u>. The threshhold concentration of ethylene causing sleepiness of carnations was about 125 ppb when blooms were exposed to the gas for 1 day at 68° F. 2 days at 50°, 4 days at 41° or 8 days at 32°. However, the threshhold was only 60 ppb when blooms were held for 3 days at 50° and was 250 ppb when held for 6 days at 32°. Vase life of blooms paralleled occurrence of sleepiness.

Sleepiness was prevented when  $10\%~\mathrm{CO}_2$  was present in an atmosphere with 250 ppb ethylene at  $68\,^\circ$  F. or when  $20\%~\mathrm{CO}_2$  was added to an atmosphere with 500 ppb ethylene at the same temperature.

Carnation blooms produce ethylene. The production is stimulated by added ethylene, except in the presence of 10% or higher  $\mathrm{CO}_2$  levels. Ethylene production is resumed when blooms are removed from a  $\mathrm{CO}_2$  atmosphere.

Packaging tests with dry ice to provide a calculated 10% CO $_2$  concentration in a polyethylene bag actually resulted in levels of 15--20% due to the added respiratory effect of the blooms. Sleepiness was effectively reduced by this technique at temperatures below  $70^\circ$  F. but above this temperature a depletion of oxygen caused injury to the blooms. (HC-34)

#### D. Postharvest Disease Control

1. <u>Carnations</u>. Carnation blooms with incipient infections of <u>Botrytis</u> <u>cinerea</u> had a 50% reduction in decay when dipped for 30 seconds in hot water at 120° F. The addition of 2-aminobutane (.01M) to the hot water reduced decay by another 50% without visible injury to the flowers. In addition to controlling decay, the hot water prolonged vase life of the blooms. (HC-34)

#### Publications - USDA and Cooperative Program

RPA 906 - CULTURE AND PROTECTION OF ORNAMENTALS AND TURF

#### Quality Maintenance in Storage

- Marousky, F. J. 1968. Influence of 8-hydroxyquinoline citrate and sucrose on vase life and quality of cut gladiolus. Proc. Fla. State. Hort. Soc. 81: 415-419. (HC-71)
- Marousky, F. J. 1969. Prolonging the vase life of cut gladiolus. Bull. No. Amer. Gladiolus Council 98: 94-101. (HC-71)

# Postharvest Physiology

Marousky, F. J. 1968. Physiological role of 8-hydroxyquinoline citrate and sucrose in extending vase life and improving quality of cut gladiolus. Proc. Fla. State Hort. Soc. 81: 409-414.

(HC-71)

AREA 12

#### VEGETABLES

Research Activity Code: 1-11-404-14-401

#### USDA and Cooperative Program

	:_	Sci	entis	t Man Y	ears,	FY 1969
Location of Intramural Work	:	Research	Prob	lem Are	a :	
	:	404	:	501	:	Total_
	:		:		:	
California	:	2.5	:	0	:	2.5
Florida	:	1.0	;	0	:	1.0
North Carolina	:	0.8	:	0	:	8.0
New Jersey	:	2.0	:	0	:	2.0
Illinois	:	1.0	:	0	:	1.0
Maryland (Beltsville)	:	3.3	:	1.0	:	4.3
Total	:	10.6	:	1.0	:	11.6

Intramural program is supplemented by extramural support representing 0.3 SMY at a State Agricultural Experiment Station.  $\underline{1}$ /

1/ RPA 404 0.3

#### Progress - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

#### A. Quality Maintenance in Storage

l. Sweetpotatoes. The amount of intercellular space in sweetpotatoes at harvest was not affected by planting dates. Irrigation with  $1\frac{1}{2}$  inches of water per day over a 10-day period did not adversely affect keeping quality of 8 varieties of sweetpotatoes, but did significantly increase the pH and reduce total acidity of the roots. Greatest changes in acidity were associated with the greatest accumulation of carbon dioxide in the roots with varieties differing slightly in their acidity responses. During storage the 8 varieties differed significantly in intercellular space, respiration rates, dry matter and water loss. Some differences were associated with skin characteristics. Stored roots subjected to increased carbon dioxide levels or held in water buffered to maintain selected pH levels, exhibited similar increases in carbon dioxide and acidity changes commonly occurring in roots previously subjected to wet soil conditions.

Increases in storage temperature increased sprouting, respiration rates, dry matter losses, and intercellular space (pithiness), whereas, decreases in relative humidity increased weight losses (through increased water losses) and volume losses but did not increase intercellular space. (HC-52)

2. Storage of mature green tomatoes in modified atmospheres. Maturegreen tomatoes held for 6 weeks in air at 55° F. developed 78% decay. Decay occurred in 100% of the fruit when it was held for an additional week at 65°. At 55° in atmospheres containing 3% oxygen with 0, 3, or 5% carbon dioxide, an average of 4% of the fruit was decayed after 6 weeks. However, 9% decay resulted after being held one additional week at 65°.

Mature-green tomatoes held at 55° were fully colored after 6 weeks in air; in a pink stage when held in 3% oxygen without carbon dioxide; and were in an advanced breaker stage when held in 3% oxygen with 3 or 5% carbon dioxide. All fruit was fully colored after ripening for 2 weeks at 60°, 65°, or 70°. Fruit ripened after a 6-week period at 55° developed the same red color as those ripened immediately at 65°.

Acidity was 37% greater in fruit ripened at 55° than when ripened at 70°, 13% greater when fruit ripened at 65° and 22% greater when ripened at 60°. Mature-green tomatoes held 6 weeks at 55° F. in 3 or 5% carbon dioxide ripened with slightly more acidity than fruits held in atmospheres with no carbon dioxide. Weight loss in tomatoes held 6 weeks at 55° F. averaged 2.1% in 14 different atmospheres with no shriveling. However, after ripening 1 week at 60°, 65° and 70°, weight losses increased to 4.3%, 4.6% and 5.7%, respectively, with slight shriveling.

Green, breaking, pink or red tomatoes inoculated with Erwinia carotovora, the soft rot bacterium, decayed more rapidly in air at 55° F. than in controlled atmospheres containing 3 or 0.25% oxygen and 5% carbon dioxide. Regardless of storage atmosphere, green tomatoes were more susceptible to soft rot decay than tomatoes at other stages of ripening. Among inoculated fruit held for 5 days at 55° F., 98% of the green tomatoes, 82% of the breaking fruit, 71% of the pink fruits and 24% of the red or fully colored fruits were decayed. The treatment of inoculated green tomato fruits with 5 to 50 ppm of ethylene gas increased both the incidence of soft rot and the ripening rate. (HC-6)

# B. Quality Maintenance during Transportation

1. Effect of modified atmosphere on lettuce in transit. Pink rib of California lettuce shipped by rail to eastern markets was significantly reduced when oxygen levels were decreased and carbon monoxide level increased in the transit atmosphere.

An unusual disorder in the form of tan to brown oval-shaped lesions was observed in railcars having 2% or higher carbon dioxide levels at destination. (HC-32)

2. Effect of loading method on transit refrigeration of western celery. Lengthwise loading of crates reduced the temperature of celery in railcars an average 3 to 5° F. Container damage and celery quality in both lengthwise and cross-wise load patterns were found to be essentially the same in preliminary tests.

In preliminary tests, pallet loads of celery in wirebound crates and waxed cartons, placed directly opposite the fans in a cold room, had half-cooling times of about 7 hours in crates and about 10 hours in cartons. (Exploratory)

#### C. Postharvest Physiology

- 1. <u>Waterscale of onions</u>. A bulk storage study indicates that neither pressure nor time in storage are significant factors in the incidence of watery scale of onion. (HC-20)
- 2. Effect of field environment on the market quality of lettuce. Sunburned lettuce heads stored 8 days at 41° F. plus 4 days at 50° showed a higher incidence of tipburn and russet spotting than those without this injury (56% vs. 45%, and 42% vs. 16%, respectively). Sunburn was confined to the cap leaf in most heads and in a few of the underlying leaves, but had no noticeable effect on pink rib.

  (HC-29)
- 3. <u>High greenhouse humidity effect on tomato fruit</u>. The principal defects on tomato fruits from plants grown in greenhouses at high relative humidity were uneven coloration, cracking, and dullness of the fruit surface. Microbial diseases were not a problem.

  (Training Program Report)

#### D. Postharvest Disease Control

- 1. Role of enzymes in the death of vegetable tissues. The sensitive viscosity assay failed to reveal collagenase activity in spores of Colletotrichum obiculare. Since commercial collagenase was also inactive, probably the collagen substrate was not sufficiently solubilized by the methods used. (HC-17)
- 2. The role of enzymes in the virulence of Erwinia carotovora. Phototrophic avirulent mutants resulted when a virulent streptomycin-resistant strain of Erwinia carotovora was treated with N-methyl-N-nitro-nitro-soguanidine. Tests indicate that the virulence in E. carotovora is associated with its production of several highly active enzymes capable of degrading or altering the permeability of the pectin, cellulose, and lecithin components of host cells. (Exploratory)

3. Control of onion neck rot. Neck rot was reduced from approximately 50% in lots without heat curing to an average of 5% in the various heat treated lots. Weight losses in storage averaged approximately 7% for the heat cured bulbs and 6% for the controls.

Lime Crest, Tutane, Thiabendazole, and Benlate, when applied to cut surfaces prior to inoculation with <u>Botrytis allii</u> spores, significantly reduced neck rot after 90 days' storage at 56° F.

Onions treated with "Shed-a-Leaf" averaged 2.3%, Herbison-treated bulbs averaged 3.5% and controls 4.2% botrytis neck rot after 3 months' storage at 56° F. Other top killers tested apparently injured tissues causing an increase in decay.

Of 20 different New York-grown onion varieties tested, "Buccaneer Harris," "Elite Harris," and "#6 Hybrid Trapp" had least decay in common storage at 56° F. (HC-16)

4. Increased effectiveness of chlorine in wash and hydrocooling water. A study of the effect of high populations of soft rot bacteria in dumping, washing, and hydrocooling water on the incidence of vegetable soft rot was made possible by the development of a specific cultural medium for the genus <a href="Erwinia">Erwinia</a>.

When carrots were washed in water containing an Erwinia population of  $1\times10^8$  per ml. the incidence of soft rot increased from 0 to 68%. Soft rot increased in radishes from 4 to 51%, in tomatoes from 2 to 26%, and in celery from 7 to 37% when they were washed in water with an Erwinia population of  $1\times10^5$  per ml. Excellent decay control was obtained when mechanically harvested green tomatoes were dumped from the harvester into a pallet box of water containing a residual chlorine concentration of 50 ppm to which the surfactant "Santomerse F-85" was added. (HC-40)

5. Identification and causes of new market and storage diseases.

Thielaviopsis basicola, the causal fungus of black mold rot, was isolated from samples of carrots grown in different western geographical locations.

Chalaropsis thielavioides also reported to cause the same disease, was not found.

A three-year study has resulted in a reevaluation of the importance of Geotrichum candidum on produce. The organism has been isolated from a total of 26 different commodities showing decay although only 14 are reported in the literature. A collection of more than 100 isolates is now being maintained in the laboratory. A Geotrichum isolated from carrot has been reported to be pathogenic to humans.

The causal agent of a hitherto undescribed bacterial stalk spotting and soft rot of Chinese chard (Bok-Choy) has been identified as a variant of Pseudomonas marginalis. The soft rot phase of the disease has caused serious losses in the Chicago market. (HC-23)

When held at  $115^{\circ}$  F., more garlic cloves developed symptoms resembling waxy breakdown at 50% relative humidity than at higher humidities at the same temperature. Two lots of California-grown garlic showed no increase in waxy breakdown when held at  $34^{\circ}$  for 18 weeks.

An apparent physiological disorder, first noted in 1961, has recently become a serious market disease of lettuce. The disease, referred to as "rib rust" first appears in January and is most serious in February and March. In 1969, approximately 90% of the February arrivals on the New York market were affected by the disorder. The symptoms are a brown discoloration of the midribs and veins of the outer head leaves.

(HC-18)

- 6. Losses in lettuce during marketing. Losses of western lettuce in the New York metropolitan area during the 1968-69 marketing year averaged 16%. Of this total loss about 7% represented the trimming loss at the ultimate use level. Decay and bruising caused most of the 4.5% loss at the wholesale and of the 4.8% loss at the retail level. Losses during the same period in the Chicago market averaged about 24% with the major portion of the loss at ultimate use. Losses at the wholesale and consumer levels were mostly physical damage whereas losses at retail were mostly from decay. (HC-19 and 24)
- 7. Decay control in California melons. The addition of 600 ppm Captan to hot water (135° F.) dips for 15, 30, or 60 seconds significantly improved control of stem-scar mold and surface mold of melons. Quality evaluations made after holding melons 7 days at 46° F. plus 3 additional days at 72° indicated that the hot water treatments increased suture browning of melons. (Exploratory)
- 8. <u>Decay control in California artichokes</u>. Preliminary tests indicate that 130° F. water dips for 60 seconds severely injured artichokes, 120° dips for 30 or 60 seconds caused browning of the bracts, but 60 second immersion in 115° water caused no injury. Dehydroacetic acid in concentrations up to 5000 ppm did not injure artichokes but did leave a visible residue. (Exploratory)
- 9. Tomato decay control. Mature-green tomatoes treated with room temperature solutions of Benlate, Thiabendazole, Botran, Geigy chemical 20092, or hot water, or hot air developed about the same amount of decay during 6 weeks at 55° F. and during ripening at 65° as did non-treated fruit. Heated (125°) solutions of Benlate and thiabendazole reduced decay development of "breaker" or "pink" tomatoes during a 6-week storage period and during ripening but not on mature-green tomatoes. (Exploratory)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- 1. Maturity in mature-green tomatoes. A new approach, that of using phototube voltage (PTV), was tested and found to be promising as a maturity sorting technique. PTV is the mean of the signal passing through 4 filters and throughout the product. Meter display of phototube response was indicated as a high value when least mature fruit was measured and as low values when more mature fruit was measured. The PTV technique sorted normal size fruit better than small size fruit. (HC-13)
- 2. Firmness of head lettuce. A low pressure bulk compression apparatus for determining head firmness was constructed. Initial tests of the system showed that boiled water was required as the compression fluid in order to obtain repeatable results. (HC-76)

#### Publications - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS & VEGETABLES

#### Quality Maintenance in Storage

- Kushman, L. J. and F. S. Wright. 1968. A new system for storing sweetpotatoes: Curing in one room and storing in another with palletized handling, overhead ventilation and trench heating and humidification. N. C. Agr. Expt. Sta. Tech. Bul. 187, 40 pp. (HC-52)
- Kushman, L. J. 1969. Inhibition of sprouting in sweetpotato by treatment with CIPC. HortScience 4(1): 61-63. (HC-52)
- Kushman, L. J. and F. S. Wright. 1969. Sweetpotato storage. USDA, Agr. Handb. 358, 35 pp. (HC-52)
- Lutz, J. M. and R. E. Hardenburg. 1969. The commercial storage of fruits, vegetables and florist and nursery stocks. USDA, Agr. Handb. 66, 94 pp. (HC-1)

#### Quality Maintenance during Transportation

- Lipton, W. J. 1968. Market quality of asparagus—effects of maturity at harvest, and of high carbon dioxide atmospheres during simulated transit. USDA, MRR 817, 7 pp. (HC-31)
- Stewart, J. K. and M. J. Ceponis. 1968. Effects of transit temperatures and modified atmospheres on market quality of lettuce shipped in nitrogen-refrigerated and mechanically-refrigerated trailers. USDA, MRR 832, 9 pp. (HC-32)

#### Postharvest Disease Control

- Beraha, L. 1968. Gray mold of green onions on the market. Plant Disease Reporter 52: 974-975. (HC-23)
- Burton, C. L. 1968. Southern corn leaf blight on sweet corn ears in transit. Plant Disease Reporter 52: 847-851. (HC-23)
- Segall, R. H. 1968. Are you making good use of chlorine? American Vegetable Grower 16(8): 14, 26. (HC-40)
- Segall, R. H. 1968. Fungicidal effectiveness of chlorine as influenced by concentration, temperature, pH, and spore exposure time. Phytopathology 58: 1412-1414. (HC-40)
- Segall, R. H. 1968. Reducing postharvest decay of tomatoes by adding a chlorine source and the surfactant Santomerse F-85 to water in field washers. Proc. Fla. State Hort. Soc. 81: 212-214. (HC-40)
- Spalding, D. H. 1969. Toxic effect of macerating action of extracts of sweetpotatoes rotted by <u>Rhizopus stolonifer</u> and its inhibition by ions. Phytopathology 59: 685-692. (HC-10)
- Wells, J. M. 1968. Growth of <u>Rhizopus stolonifer</u> in low-oxygen atmospheres and production of pectic and cellulolytic enzymes. Phytopathology 58: 1598-1602. (HC-30)

#### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

- Anonymous. Light technique adapted to sorting green tomatoes.

  Agricultural Research 18(8): 3. (HC-13)
- Kushman, L. J., D. T. Pope, and J. A. Warren. 1968. A rapid method of estimating dry matter content of sweetpotatoes. Proc. Amer. Soc. Hort. Sci. 92: 814-822. (HC-52)
- Worthington, J. T. 1969. Maturity sorting of green tomatoes. United Fresh Fruit & Vegetable Association 1969 Yearbook: 283-286. (HC-13)
- Worthington, J. T., R. W. Penney and J. N. Yeatman. 1969. Evaluation of light source and temperature on tomato color development during ripening. HortScience 4(1): 64-65. (HC-13)

# PROGRESS REPORT OF THE STORED-PRODUCT INSECTS RESEARCH BRANCH MARKET QUALITY RESEARCH DIVISION

# MARKET QUALITY RESEARCH DIVISION AGRICULTURAL RESEARCH SERVICE

#### AREA 2

#### DAIRY PRODUCTS

Research Activity Code: 2-41-412-14-609

#### USDA and Cooperative Program

: Scientist Man-Years	F.Y. 1969
: Research Problem Area	3 :
: 408	: Total
•	:
: 1.9	: 1.9
. 10	. 10
· 1.7	: 1.9

Intramural program is supplemented by extramural support representing 0.1 SMY at a State Agricultural Experiment Station.

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

- 1. Stored-Product Mite Studies. Radio frequency treatment of dogfood infested with the mite Tyrophagus putrescentiae has indicated mite control was obtained when temperatures of 137° F or higher were reached during a 21-second exposure. (SP 7-2)
- 2. Transmission of Salmonella by insects. An insect-mounting procedure was devised that facilitated quantitative studies of feces with a minimal possibility of cross-contamination. By this method it was observed that 10 cells of Salmonella were required for passage through the intestinal tract of Dermestes maculatus. Multiple doses of this magnitude were necessary to initiate intestinal infection. It is doubtful that D. maculatus is involved significantly in the dissemination of Salmonella. (SP 7-3)

3. Survey of Stored-Product Insects. The dermestid, Reesa vespulae (Milliron), has been found in Wisconsin for the first time. This insect reproduces by parthenogenesis. Only females have been observed. (SP 7-4)

## B. Biological and Physical Control

- 1. Insect Response to Pheromones. Two of the pheromones of Trogoderma inclusum have been identified as 14-methyl-cis-8-hexadecen-1-ol and methyl-14-methyl-cis-8-hexadecenoate. T. glabrum responds strongly to the trans isomer of the alcohol compound. Evidence for sex pheromones has been demonstrated in T. simplex, T. variabile (=T. parabile), T. sternale, and T. grassmani and each responds to the cis alcohol T. inclusum synthetic compound. It appears the sex pheromones of several Trogoderma species may be similar. The crude sex pheromone of the cigarette beetle has been used successfully to lure males from a mixed population into a trap. Black carpet beetle females appear to be monogamous. (SP 7-1)
- 2. Microbial Symbionts Protozoa. The gregarine, Pyxinia frenzeli, from the alimentary tract of the black carpet beetle is mildly pathogenic under dietary stress conditions. The nature of attachment by the protozoa to the host midgut wall was established. The insect host larva loses its midgut epithelium with each larval molt so infection of this gregarine must begin anew with ingestion of sporocysts by each instar. The protozoa have never been found parasitizing the midgut of the adult black carpet beetle, however, the newly emerged adults carry the protozoa in a blind sac which is the intact remnant of the larval midgut. Gregarine-infected larvae lose weight almost twice as rapidly as noninfected larvae under starvation conditions and may aid in suppressing field populations of the insect. (SP 5 (A8475))
- 3. Microbial Symbionts Sorbic Acid. Mated pairs of the black carpet beetle and Trogoderma variabile (=T. parabile) developing from sorbic acid treatments produced progeny unable to survive the first larval instar. This delayed, but acute effect of sorbic acid, a common food ingredient, represents the most practical immediate result obtained from this series of studies. (SP 5(A8475))
- 4. Attractants of <u>Trogoderma spp</u>. Four compounds, each active by itself, comprise the attractant of <u>T. inclusum</u>. Two of the compounds have been identified as 14-methyl-cis-8-hexadecen-1-ol and methyl 14-methyl-cis-8-hexadecenoate and have been synthesized. The <u>trans</u> isomers were also synthesized. Synthesis work is proceeding in an effort to obtain adequate supplies for biological studies. (SP 1 (C9500))

- 5. Synthesis of Megatomoic Acid. Synthesis of megatomoic acid, the attractant for Attagenus megatoma has been completed. A 1-gram sample containing about 35% megatomoic acid and 71-grams of 50% megatomoic acid have been prepared. (SP 73 (C9532))
- 6. Constituents of Cheese that Attract Mites. The neutral compounds responsible for the cheese-mite attractancy were isolated. The major attractive factors were characterized as heptan-2-one, octan-2-one, nonan-2-one, undecan-2-one, tridecan-2-one and a nonen-2-one. Significant attractivity was shown only when the individual compounds were combined. (SP 42 (All-MQ-3))

# C. Improved Insecticidal Control

1. Residual Insect Control Treatments. Twenty different building material surfaces have been studied to determine the residual toxicity of malathion. At 1 gm/m² malathion was toxic for 27 weeks on glass and was essentially nontoxic on cement. Fenthion, Dursban®, Baygon®, and bromophos were comparable to malathion on similar surfaces.

The residual effectiveness of malathion could be improved by covering cement surfaces with coating agents such as talc or certain types of paints prior to applying the malathion. (SP 6 (A9391))

# Publications - USDA and Cooperative Program

- Dunkel, Florence V. and Boush, G. Mallory. 1968. Biology of the gregarine Pyxinia frenzeli in the black carpet beetle, Attagemus megatoma. Jour. Invertebrate Path. 11(2):281-88. (SP-5)
- Julseth, R.M., Felix, Jacob K., Burkholder, Wendell E., and Deibel, R.H. 1969. Experimental transmission of Enterobacteriaceae by insects. I. Fate of Salmonella fed to the hide beetle Dermestes maculatus and a novel method for mounting insects. Applied Microbiology 17(5):710-713. (SP-7)
- Slominski, J.W., Gojmerac, W.L. and Burkholder, W.E. 1969. Oils: Their effect on black carpet beetle larvae when applied to different types of surfaces. Jour. Econ. Entomol. 62(2):507-508. (SP-6)

#### AREA 3

#### DECIDUOUS FRUITS AND TREE NUTS

Research Activity Codes: 1-13-404-14-605

1-15-404-14-606

#### USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1969				
Location of Intramural Work	Research Proble	em Area : Total			
Dried Fruits California	: : : 3.2	: : : 3.2			
Tree Nuts California	1.8	1.8			
Total	5.0	: 5.0			

Intramural program is supplemented by extramural support representing 0.1 SMY at a State Agricultural Experiment Station.

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

- 1. Biology, Ecology, and Behavior of Oryzaephilus spp. O. mercator does better than O. surinamensis on rolled oats, almonds, and raisins at 30°C and 20% R.H. Neither species completed development on prunes. Both species had longer life cycles at 30°C and 20% R.H. than at either 30°C and 50% R.H. or 35°C and 80% R.H. (SP 46-1)
- 2. Identifying Characters of Immature Carpophilus. A total of 201 accessions of nitidulid beetles were obtained from various areas in the southern United States and Hawaii. Approximately 150 laboratory cultures were established. Microslide preparations of the adults of 35 species of Carpophilus were made. Illustrations of adults were prepared for 12 species of Carpophilus, and one species each of Coleopterus, Epuraea, Stelidota, and Urophorus. Ninety-two microslide preparations of larvae were made. Preliminary studies showed that good larval characteristics for differentiation were present. (SP 37 (A9710))

3. Ecology of Mites on Dried Fruits and Herbs. Two species of mites Tyrophagus putrescentiae and Rhizoglyphus echinopus, developed and produced eggs when feeding on almost all herbs. Two other species of the genus Acarus and Carpoglyphus lactis fed only on a few herbs. Herb fruits and seeds were more infested than were leaves, flowers, and roots. (SP 44 (E21-MQ-1))

# B. Improved Insecticidal Control

- 1. <u>Drosophila Control</u>. In tests with an automated dichlorvos aerosol system in a wine cellar 95% of caged drosophila and 56% of caged dried fruit beetles were killed with 1-hr. exposures. (SP 46-5)
- 2. Funigation of Dried Fruits. Ethyl formate applied at the rate of 10 ml. per 30-lb. carton of raisins was effective against various stages of the Indian-meal moth, saw-toothed grain beetle, merchant grain beetle, and red flour beetle. Few larvae of Trogoderma variabile (=T. parabile) survived. Average concentrations of ethyl formate in mg. per 1. at the end of 5 min. were 108; 10 min., 180.7; and 20 min., 75. (SP 46-9)
- 3. Protectants for Dried Fruits in Commercial Storage. Insects not found extensively in raisins for many years are appearing in raisins treated by the malathion-treated drying tray method. They are the black raisin moth, Ephestoides gilvescentella, and the tobacco moth, Ephestia elutella. Malathion on the raisins is at the 5 p.p.m. level. The moths are causing little damage. (SP 46-10)
- 4. Effect of Malathion on Reproduction of Plodia. A laboratory population of Indian-meal moth was found to eliminate almond moth when both were placed (as eggs) in untreated or in 1 p.p.m. malathion-treated almond nutmeats. When 2 or 3 p.p.m. of malathion was used on almonds the Indian-meal moth was eliminated and some almond moth survived. Diel cycle of egg production by Indian-meal moth, maintained under ambient conditions in a warehouse, occurred at approximately 11 a.m. and 8 p.m. Pacific Daylight Time (PDT) each day. The smaller peak (11 a.m.) may be correlated with high incident radiation within the warehouse. At Fresno, the highest incident solar radiation normally occurs at 11 a.m. (PDT). Peak air temperature in the warehouse was at 5 p.m. (PDT). This may have influenced the highest peak of egg production (8 p.m.). These findings indicate that use of malathion at low dosages (2 and 3 p.p.m.) may effect a change in most predominant species in stored almonds. (SP 47-2)
- 5. Protectants for Tree Nuts. The quality of almonds has been maintained for 9 months by treating them with malathion at rates of 4, 8, and 16 p.p.m. in laboratory tests. At 8 months, however, Indian-meal moths were present in numbers sufficient to eventually become a problem. Normally, untreated nuts are completely consumed by insects in 6 months. (SP 47-3)

6. Fumigation of Stored Tree Nuts. Inshell Nonpareil almonds, Hartley walnuts, and raw and roasted peanuts were fumigated with phosphine evolving from aluminum phosphide tablets at the rate of 50 per 1,000 cu. ft. Exposure was for 6 days in a chamber. Residues were less than 0.004 p.p.m. and no off-odor or off-flavor developed. (SP 47-4)

## C. Insect-Resistant Packaging

1. Cartons for Dried Fruits. Raisins in  $l\frac{1}{2}$ -ounce cartons made of bleached kraft board and coated on both sides with 1-mil polyethylene were not infested after 3-months' exposure to insects. However, raisins in 15-oz. cartons of the same construction became infested. These were examined for structural defects. Minor changes corrected the defect and the industry is now using the improved cartons. Another carton, polyethylene-coated with hot-melt seal, shows promise. An automatic leak detector is being developed by means of which the degree of insect infestation may be correlated with tightness of the carton. (SP 46-6)

# Publications - USDA and Cooperative Program

- Boczek, Jan, and Czajkowska, Barbara. 1968. The effect of antimicrobial agents and antibiotics on some stored products mites (Acaroidae). Roczniki Nank Rolniczych 93(4):597-612. (SP-44)
- Curtis, Charles E. 1968. Research activities at the Stored-Product Insects Research Branch. Proc. Eighth Ann. Res. Conf., Dried Fruit Indus. Res. Advisory Com. (SP-46)
- Lovitt, Anthony E., and Soderstrom, Edwin L. 1968. Predation on Indianmeal moth eggs by <u>Liposcelis</u> bostrychophilus. Jour. Econ. Ent. 61(5):1444-1445. (SP-46)
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AREA 4

GRAIN

Research Activity Codes: 1-1-408-14-601

1-4-408-14-602

## USDA and Cooperative Program

	:	Scientist Man-Years F.Y		1969
Location of Intramural Work	:_	Research Problem Area	:	
	:	408	:	Total
	•		:	
Kansas	:	7.0	•	7.0
Georgia	•	0.2	:	0.2
	:		:	
	•		•	
Total	:	7.2	:	7.2
	:	_	:	

Intramural program is supplemented by extramural support representing (a) 1.0 SMY at State Agricultural Experiment Stations and (b) P. L. 480 funds in one country representing 137,110 U. S. dollars equivalent.

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

Biology of the Angoumois Grain Moth. The greatest variability of Angoumois grain moth pupal periods was among those reared in whole wheat kernels, a range of 7 to 12 days. Most of the individual variation in developmental periods of moths reared on a given diet occurred during the larval period. Individuals reared in whole kernels showed the shortest mean larval period (28.2 days). Those reared on certain meal diets of endosperm-germ mixtures showed less larval period variation. The longest mean larval period in any diet tested was for those reared in purified endosperm pellets (138.5 days). Moths reared in finely ground meals exhibited longer larval periods than those reared in coarser meals. Most larvae not reaching the adult stage died while still very small. If they were large enough for X-ray detection, they usually completed development. Larvae were found to live in purified endosperm pellets for as long as 252 days; however, none having a larval period longer than 239 days pupated and none emerged that had a larval period exceeding 180 days. Extensive data have been collected and assessed, and chi-square tests confirm that the red-eyed mutant is autosomal recessive. (SP-22(A8404))

- 2. Water Regulation Physiology and Stress. Radiotracer studies on water absorption by the hindgut of yellow mealworm larvae showed the effect of solute concentration on water transport. Tritiated water solutions containing different molarities of the disaccharide trehalose were profused into the in vivo rectal preparation. Water was rapidly taken up by the rectal epithelium, even from solutions with high solute concentrations. Water containing no solute was absorbed to a greater extent than when solute was present. There was considerable influx as well as efflux from the rectal lumen, suggesting a recycling mechanism of water transport. Tracheal ventilation considerably affected quantity of original lumen water absorbed by individual cockroaches even though such variables as age, sex, temperature, etc. were controlled. A tentative correlation was made between the degree of activity shown by individual insects and the quantity of water absorbed. Faster absorption was associated with more activity. Severing rectal nerves had no effect on the quantity of water absorbed or water diffusion into the lumen. Cutting all tracheal connections reduced absorption, supporting the observation that inactive insects absorbed water at a lower rate than those ventilating. Preliminary work to establish biochemical differences between two strains of yellow mealworms have shown differences in rates of growth and development under dry and wet conditions. Uniformly labeled C14-glucose when injected into individual mealworms was rapidly metabolized. (SP-27(A9381))
- 3. Distribution of Hazardous Microorganisms. The effect of Salmonella montevideo on the development of Sitophilus oryzae was determined. Adult rice weevils in S. montevideo-contaminated wheat had lower mortality rates than did weevils in clean wheat. The average number of progeny per female parent was higher in the wheat infected with S. montevideo (137) than in the clean wheat (103.5). Weevils in contaminated wheat experienced shorter emergence periods than weevils in clean wheat. S. granarius, S. oryzae, Rhyzopertha dominica, Tenebroides mauritanicus, Oryzaephilus surinamensis, Cryptolestes pusillus, and Tribolium castaneum were able to pick up S. montevideo in 1 week. All species were able to contaminate clean wheat.

Progeny of S. oryzae, O. surinamensis, and T. castaneum reared in S. montevideo-contaminated wheat, carried the bacteria into clean wheat. A direct relationship existed between the length of time weevils were exposed to contaminated wheat and how long they retained the bacteria. All ages and all life stages of the Indian-meal moth were found to pick up and carry S. montevideo. The larvae passed the contamination into the pupal stage then to the adult which in turn laid contaminated eggs. One species of Aspergillus flavus produced toxin above 229.9 p.p.b. while growing in moist yellow corn at 85°F. The same microorganism growing at lower temperatures produced a variety of toxins at lower concentrations. Other tests have shown that certain stored-grain insects will carry Aspergillus flavus from inoculated to sterile grains resulting in toxin development. (SP-28(A9403))

4. Effect of Dockage on Grain Insects. Habitational preference and fecundity studies were completed on rice weevil and lesser grain borer. The results indicate that adults of both species have a slight preference for clean wheat over that containing 1 to 4 percent dockage. Twenty-three percent of the rice weevil progeny developed in the clean wheat compared to 18.2 percent in wheat containing 4-percent dockage. As for the lesser grain borer, 28.6 percent of the progeny produced were found in the clean wheat compared with 13.9 percent in the wheat containing 4-percent dockage. (SP-49-5)

An exploratory study was conducted to determine whether it would be possible to conduct a full scale test with the flat grain beetle. All test insects were recovered from the clean wheat but only 36 percent of the flat grain beetles were recovered from the grain containing  $4\frac{1}{2}$ -percent dockage. Only 74 progeny developed from 250 adults which were in the clean grain, and only 51 developed in the wheat with  $4\frac{1}{2}$ -percent dockage. (SP-49-5)

- 5. Ecology of Grain Insects. Grain that had been used previously to rear the rice weevil had an excitant effect on several different field collected strains of the same species. This resulted in production of a greater number of progeny than in clean grain. (SP-49-7)
- 6. Feeding Habits of Insects on Soybeans. Ten generations of Indianmeal moth were reared on soybeans. The life cycle, weight, etc., were compared with those reared in grains. Larvae damaged whole beans. Cadelle infestations developed in clean, whole beans. (SP-50-7)
- 7. Insects in Bulgur. Tests were conducted, using several species of insects, in bulgur and wheat held at relative atmospheric humidities of 71-, 58-, and 43-percent. With the exception of the black carpet beetle, the following test insects showed longer developmental periods in bulgur than in wheat at all moisture levels: Indian-meal moth, confused flour beetle, saw-toothed grain beetle, flat grain beetle, Trogoderma variabile(=T. parabile), rice weevil, and lesser grain borer. There appeared to be little difference for the black carpet beetle. (SP-25(A9353)
- 8. Insect Survey in Uttar Pradesh. Uttar Pradesh was divided into 6 zones for the purpose of stored-grain insect surveys which could be utilized in a program of control. The type of storages and climatic conditions vary widely from one zone to another. It was found that Sitophilus oryzae and Rhyzopertha dominica were present in empty stores of all the regions. Trogoderma granarium was found in 4 of the 6 regions. (SP-68(A7-MQ-15))

- B. Biological and Physical Control
- 1. <u>Insect-Resistant Corn Varieties</u> Data have been collected from 176 varieties of corn evaluated for maize weevil resistance.(SP-52-2)
- 2. Resistance of Varieties of Grain Sorghum. The resistance of sorghum varieties to infestation by both Sitophilus oryzae and S. zeamais is significantly affected by varietal resistance factors developed in the grain through 6 generations. (SP-20(G9177))
- 3. <u>Insect-Resistant Wheat Varieties</u>. Of 152 varieties of wheat tested for resistance to rice weevil, <u>Sitophilus oryzae</u>, only one variety, E4007, was found to be comparatively resistant. Eleven of 170 varieties of wheat tested were resistant to lesser grain borer, <u>Rhyzopertha dominica</u>. (SP-38(A7-MQ-1))
- 4. Attractants of Khapra Beetle. Fatty acid esters have been identified from both sexes of Trogoderma granarium. After identification, each of these esters was synthesized and bioassay was carried out. T. granarium males and females were attracted by the esters and Tribolium castaneum were repelled by the same compounds. (SP-41 (AIO-MQ-11))
- 5. Diapause in Trogoderma. Extensive studies are in progress to isolate and characterize the biologically active principle present in fecal lipid extracts. Trogoderma larvae reared singly in combinations with wheat flour and fecal pellets were observed to determine the effect of fecal material and its constituents on diapause. It was observed that the larvae entered diapause in significantly increased percentages in cultures with added larval fecal pellets. A certain percentage of diapause could be induced by addition of extracted fecal lipids into the food. More larvae entered diapause when the food contained greater percentages of fecal lipids. The addition of fecal pellets at a concentration of 50% of the total diet substrate had no effect in inducing diapause when the food available was 100 mg. per larva. The effect of added fecal pellets began to manifest itself when the amount of food was reduced to 30 mg. per larva. (SP-39(A7-MQ-3))

The study of factors related to induction as well as termination of larval diapause was continued. It appears that at least 2 chemically distinct groups of materials are involved in the induction of diapause in Trogoderma granarium, each at a different stage of larval development. (SP-41(AlO-MQ-11))

6. Pathogens. Biopreparations containing the bacterium, Bacillus thuringiensis, and the fungus, Beauveria bassiana, were tested for control of insect pests of cereal grain and flour. Some of the test insects were successfully controlled by these pathogens, but the greater humidity which is favorable for some microorganisms can spoil the quality of the treated commodity. (SP-45(E30-MQ-1))

7. Effect of Temperature and Humidity. The creation of unfavorable environmental conditions was accomplished to effect control of certain stored-grain insect pests. Humidity of environment and food greatly influenced the resistance of Sitophilus granarius adults to moderately low temperatures. Their larvae were more susceptible than were the adults. In the investigations of Sitotroga cerealella and Plodia interpunctella, it was found that when the eggs were exposed to low temperatures development was prolonged and the number of hatched larvae from such eggs decreased. (SP-45 (E30-MQ-1))

# C. <u>Improved Insecticidal Control</u>

- 1. <u>Dichlorvos Aerosol in Boxcars</u>. Six ounces of a pressurized aerosol containing 6.5-percent dichlorvos produced complete mortality of test insects in boxcars after an exposure of 15 minutes. Kill was more rapid near the floor than near the roof. (SP-49-1)
- 2. Candidate Protectants on Wheat. In competitive food acceptability tests, rice weevils entered samples of untreated wheat and wheat treated with Gardona, 5-percent malathion granular carbon, and malathion ULV concentrate in about equal numbers. Wheat treated with a diatomite, (Kenite 2-I) and a silica aerogel (Cab-O-Sil) remained highly repellent for 30 months. The insecticidal effectiveness of these materials on 10- and 12-% moisture wheat remained unchanged on undisturbed lots for 24 months.

Laboratory studies of the effectiveness of surface applications of candidate insecticidal formulations for moth control indicate that Indian-meal moth adults can penetrate the surface layers of wheat containing as much as 160 p.p.m. malathion and deposit fertile eggs. The larvae completed development below the treated surface areas and most of these migrated up through the treated wheat on the surface; some fed and spun webs on the surface while others re-entered the wheat below the surface. Some mortality was noted in the migrating larvae and pupae; however, normal pupae were evident and many adults emerged.

Toxicity studies conducted 1 month after treatment show that a dosage of 5 p.p.m. of Bay 77488 was effective against rice weevils, red and confused flour beetles, and lesser grain borers. (SP-49-2)

3. Dichlorvos for Moth Control. Resin pellets containing 18.6% dichlorvos controlled Indian-meal moth infestations when used at rates of 105-157.5 gm. per 1,000 cu. ft. Surface applications of dichlorvos in deobase oil (80 p.p.m. in the top 1-in. of wheat) gave excellent kills for 1 month. Dichlorvos in water did not perform as well. In simulated 3,250-bu. bins, tests showed that Indian-meal moth adults were killed 24 hr. after emergence for 18 weeks with one resin strip (105 gm. per 1,000 cu. ft.). Less than 0.3 p.p.m. dichlorvos was found in the top 2 in. of wheat after 18 weeks. (SP 49-3)

- 4. Malathion on Farm-Stored Wheat. Farmer applications of malathion were improved in a test series to give more uniform distributions. Most of the wheat contained 2 to 5 p.p.m. After 4 months, 97% of the bins were insect-free except for infestations of Indian-meal moth. About 3% were infested with flat grain beetles. (SP-49-6)
- 5. Forced Distribution of Fumigants. Component relationships were determined during penetration of ethylene dichloride:carbon tetrachloride (75:25) in small metal towers of wheat at 25° C. and 12.5% moisture. Gas samples obtained during the initial surge of gas through the wheat mass showed component ratios of 1.1 part EDC to 1 part CCl4 at the 1-ft. level, 1 to 4 at the 3-ft. level, and 1 to 16 at the 5-ft. level. The ratios remained unchanged up to and including the period in which peak concentrations were obtained at each sampling level. (SP-49-8)
- 6. Phosphine Fumigation of Farm-Stored Wheat. Aluminum phosphide tablets at the rate of 90 per 1,000 bu. were probed into 5 bins of wheat stored on farms. Post-fumigation sampling revealed that the wheat remained nearly free of insects throughout the fall and winter months. (SP-49-9)
- 7. Malathion Aerosol Treatment of Corn. Malathion aerosol applied to 3,250 bu. of shelled corn resulted in 17 p.p.m. on the surface to 0.7 p.p.m. in the center. (SP-50-1)
- 8. Phosphine for Pink Bollworm. Phosphine readily penetrated the several types of bags used for packaging cottonseed. Phosphine was retained almost as well by 1.5 mil as by 6-mil polyethylene sheeting used as a cover for stacks of bags during fumigation. Bagged cottonseed and bulk, fuzzy cotton-seed were fumigated under 1.5 mil poly tarps to establish the dosage rate of 60 aluminum phosphide tablets per 1,000 cu. ft. with an exposure period of 120 hr. for quarantine control of pink bollworm. This treatment has been adopted by the Plant Pest Control Division. (SP-50-2)
- 9. Candidate Protectants for Corn. Tests with malathion at 5% in granular carbon, and in low-volume emulsion and ultra-low-volume spray applications to corn were slightly more effective than the standard malathion application. (SP-50-4)
- 10. Dichlorvos Strips to Control Moths in Soybeans. Dichlorvos resin strips effectively controlled Indian-Meal moth infestations in soybeans in pilot scale laboratory tests. The strips were used at a dosage of 1 and  $1\frac{1}{2}$  per 1,000 cu. ft. of overspace in bins that were 7.5 ft. in diameter and were scale models of the upper part of regular 3,250-bu. metal bins. (SP-50)
- 11. Dichlorvos-in-Oil as Top Dressing for Soybeans. Dichlorvos in oil gave excellent kill of moths in soybeans. Residues of 0.3 p.p.m. and less were recovered over a period of 16 weeks. (SP-50-5)

- 12. Funigant Evaluations for Soybean Insects. Eggs, larvae, and pupae of the Indian-meal moth were exposed to funigants. Eggs were resistant to phosphine, but susceptible to chloropicrin. The four funigants ranked in overall order of effectiveness were chloropicrin, 75:25, phosphine, and 80:20. Tests indicated that 75:25 components were sorbed less in soybeans than in corn, wheat, or sorghum. (SP-50-6)
- 13. Dichlorvos for Moths in Small Bins. Screening tests with different formulations of dichlorvos applied to small storage chembers containing shelled corn infested with all life stages of the Indian-meal moth showed that polyvinyl chloride resin impregnated with dichlorvos gave the best overall control of the moth population. In tests with a dosage of 1.0 strip (18.6% dichlorvos) per 1,000 cu. ft. of overspace conducted in bins 7-ft., 5-in. in diameter, all adult moths emerging from infested material and all moths introduced from laboratory cultures were killed and the corn remained free of an infestation during the last 12 weeks of a 16-week observation and study period. Residues of 0.3 p.p.m. or less of dichlorvos have been found on the top 2 in. of corn following continuous exposure to vapor emitted by the strip. (SP-50-8)
- l4. Dichlorvos Strips for Moths in Grain Sorghum. Resin strips impregnated with dichlorvos were installed at a dosage of l strip per 1,000 cu. ft. of overspace in bins containing grain sorghum. Fertilized moths introduced in the overspace were able to deposit viable eggs before being killed by the vapor emitted by the strip, but only a few  $F_1$  progeny adults emerged as a result of these depositions. All adults emerging in the bins were killed before they were able to deposit viable eggs. (SP-51-2)
- 15. Inert Dusts. The influence of 3 different relative air humidities upon the effect of 19 different inert dusts on 6 species of stored-grain insects has been investigated. Generally, the granary and rice weevils were most resistant followed by the flour beetles, bean beetle, saw-toothed grain beetle, and Siamese grain beetle. In studies to determine the correlation between individual inert dusts and the loss of moisture extracted from insect bodies, the bean weevil died when it lost 20 to 30% of its body weight. (SP-45(E30-MQ-1))

# D. Fate and Effect of Residues

1. Fate of Malathion on Grain Sorghum. A cleanup procedure has been developed and published that permits the use of the electron capture detector for the gas chromatographic analysis of organophosphorous insecticide residues.

Procedures for extraction, cleanup, and determination of malathion and its hydrolytic products in grain sorghum by gas liquid chromatography have been developed. Thin layer chromatographic systems for resolving the components of a mixture of malathion and its hydrolytic products have been established.

Esterase inhibition of common stored-grain insects by malathion and its oxygen analog, malaxon, employing agar gel and electrophoretic methods continued to be investigated. (SP-34(A9706))

## Publications - USDA and Cooperative Program

- Alumot (Olomucki), Eugenia, and Mandel, E. 1968. Field test for the detection of halogenated hydrocarbon residues in fumigated whole cereals. Jour. Stored-Prod. Res. 4(1):83-85. (AlO-AMS-4)
- Alumot (Olomucki), Eugenia, Nachtomi, Edna, Kempenich-Pinto, Ora, Mandel, E., and Schindler, H. 1968. The effect of ethylene dibromide in feed on the growth, sexual development and fertility of chickens. Poultry Sci. 47(6):1979-1985. (Alo-AMS-4)
- Bell, K. O., and Mills, R. B. 1968. A comparative study of the biology of the Angoumois grain moth reared in wheat kernels and in various diets composed of purified milled wheat fraction. Abstract in Proc. North Central Br. ESA 23(1):36. (SP-22(A8404))
- Boles, H. P., and Mahany, P. G. 1967. A method of determining rate and time of egg deposition by insects. Jour. Econ. Ent. 60(6):1763. (SP-49)
- Husted, Stanley R., and Mills, Robert B. 1969. Interspecific kernel-entry behavior and competition among larvae of the Angoumois grain moth. Trans. Kans. Acad. Sci. 72(2). Ph. D. thesis. (SP-22(A8404))
- Kadoum, A. M. 1968. Cleanup procedure for water, soil, animal and plant extracts for the use of electron capture detector in the gas chromatographic analysis of organophosphorus insecticide residues. Bul. Environmental Contamination and Toxicol. 3(4):247-253. (SP-34(A9706))
- Karnavar, G. K. 1967. Studies on the biology of the khapra beetle, <u>Trogoderma granarium</u> Everts under laboratory conditions, with emphasis on diapause. Jour. Anim. Morph. and Physiol. 14(2):205-215. (SP-39 (A7-MQ-3))
- Karnavar, G. K., and Nair, K. S. S. 1968. Observation on the cytology and cytochemistry of the fat body of normal and diapausing larvae of Trogoderma granarium Everts (Col., Dermestidae). Jour. Anim. Morph. and Physiol. 15(1, 2):153-161. (SP-39(A7-MQ-3))
- Karnavar, G. K., and Nair, K. S. S. 1969. Changes in body weight, fat, glycogen, and protein during diapause of Trogoderma granarium. Jour. Insect Physiol. 15(1):95-103. (SP-39(A7-MQ-3))
- La Hue, Delmon W. 1969. Evaluation of several formulations of malathion as a protectant of grain sorghum against insects--in small bins. Mktg. Res. Rpt. 828, 19 pp. (SP-51)

- Mills, Robert B., and Daniels, Eddie K. 1968. Susceptibility of bulgur to several species of external-feeding stored-product insects. Bul. Ent. Soc. Amer. 14(3):212. (SP-25(A9353))
- Mills, Robert B., and Wilbur, Donald A. 1969. Insect infestations in bulgur. Proc. North Central Br. Ent. Soc. Amer. 24(1):20. (SP-22(A8404))
- Nachtomi, Edna, Alumot, Eugenia, and Bondi, A. 1968. Biochemical changes in organs of chicks and rats poisoned with ethylene dibromide and carbon tetrachloride. Israel Jour. Chem. 6(5):803-811. (AlO-AMS-4)
- Nair, K. S. S., and Karnavar, G. K. 1968. A cytological study of changes in the fat body of <u>Trogoderma granarium</u> during metamorphosis, with special reference to the proteinaceous globules. Jour. Insect Physiol. 14(11):1651-1659. (SP-39(A7-MQ-3))
- Robinson, Robert R., and Mills, Robert B. 1968. Susceptibility of bulgur to infestation by internal-feeding stored-grain insects. Bul. Ent. Soc. Amer. 14(3):212. (SP-25(A9353))
- Yinon, U. 1968. Mass breeding of the khapra beetle. Jour. Econ. Ent. 61(6):1738-1739. (SP-41(AlO-MQ-11))
- Yinon, U., and Shulov, A. 1967. A bioassay of the pheromone of <u>Trogoderma</u> granarium males as an attractant for both sexes of the species.

  Ent. Expt. and Appl. 10(3, 4):453-462. (AlO-AMS-11)
- Yinon, U., and Shulov, A. 1969. Response of some stored-product insects to <u>Trogoderma granarium</u> pheromones. Ann. Ent. Soc. Amer. 62(1): 172-175. (SP-41(AlO-MQ-11))

### AREA 4a

RICE

Research Activity Code: 1-6-408-14-604

# USDA and Cooperative Program

Location of Intramural Work	Scientist Man-Years F.Y. 1969 Research Problem Area:
	: 408 : Total
Texas	1.0
Total	1.0

Intramural program is supplemented by extramural support representing P.L. 480 funds in one country representing 11,748 U.S. dollars equivalent.

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Mutrition

- 1. Development of Insect Populations. In laboratory tests rice weevil population development in rough rice appears to be a direct result of the percentage of broken kernels which varies between varieties. (SP 48-2)
- 2. Constituents of Rice that Attract Insects. Both water-partitioning (AW) fraction and ether-partitioning (AE) fraction of an ether extract of rice gave moderate to strong attractancy to the rice weevil. Major attractancy was with the AW fraction which is a complex mixture. Fractionalization is in progress. (SP 42 (All-MQ-3))

# B. <u>Improved Insecticidal Control</u>

1. Malathion Protectant Studies. Preliminary results of small-bin tests of rough rice treated with 1.0, 1.5, and 2.0 pints of malathion per 1,000 bushels reveal that respective residues after 90 days were 4, 7, and 8 p.p.m. on rough rice; 15, 22, and 31 p.p.m. on hulls; and 13, 18, and 25 p.p.m. on bran. Levels on milled rice did not exceed 0.32 p.p.m. at

- any sampling period for any treatment. Infestations of rice weevil, lesser grain borer, or confused flour beetle did not develop in any of the treated rice during the 90-day period. (SP 48-1)
- 2. Dichlorvos Protectant Studies. Dichlorvos applied to rough rice in laboratory tests at the rate of 10 p.p.m. killed all rice weevils, flat grain beetles, and Angoumois grain moths. At 5 p.p.m., rice weevil mortality was only 90% but no progeny developed. Dichlorvos, at 20 p.p.m., was effective for 3 to 6 weeks in protecting rough rice of 12.5% moisture content against rice weevil and lesser grain borer attack but not against the confused flour beetle. At 13.5% moisture, protection was less than 3 weeks. (SP 48-3)

# Publications - USDA and Cooperative Program

- McGaughey, W. H. 1968. Stored-rice insects investigations. The Rice Journal 71(7):32. (SP-48)
- Cogburn, R. R. 1967. Fumigation of bucket elevators with phosphine gas to control rice weevil and red flour beetle adults. J. Econ. Entomol. 60(5):1485-1486. (MQ 1-19)
- Cogburn, R. R. 1967. Laboratory tests of five new insecticides as protectants for stored rough rice. J. Econ. Entomol 60(5):1286-1289. (MQ 1-19)
- USDA, ARS Information. 1968. Clues to rice weevil attractant. Agr. Res. 17(4):15-16.(SP-42 (All-MQ-3))

## AREA 6

#### OILSEEDS AND PEANUTS

Research Activity Code: 1-18-408-14-607

# USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1969				
Location of Intramural Work	: Research Problem Area	•			
	: 408	: Total			
	:	:			
Georgia	: 1.8	: 1.8			
	•	:			
	:	:			
Total	: 1.8	: 1.8			
	:	:			

Intramural program is supplemented by extramural support representing P.L. 480 funds in two countries representing 173,269 U.S. dollars equivalent.

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

- 1. Insect Detection in Operative Shelling Plants. Peanuts were exposed in 1-pint cups in 11 shelling plants. In the spring sampling, almond moths, corn sap beetles, red flour beetles, merchant grain beetles, and Indianmeal moths accounted for 50, 18, 16, 7, and 1 percent of the catch in the cups that were infested. Eleven percent of the cups were infested. During early summer, the Indian-meal moth population increased to 27 percent of the total catch when 60 percent of the cups of peanuts became infested. (SP 53-3)
- 2. Effect of Processing on Infestation. Farmers stock peanuts previously infested with insects were shelled in an experimental pilot shelling plant to determine the destination of insects during the processing. Approximately 82 percent of the live insects were removed from the farmers stock peanuts as they were cleaned prior to shelling. Another 16 percent were removed with the loose-shelled kernels and oilstock peanuts destined for crushing. (SP 53-4)

3. Insect Detection in Closed Shelling Plants. Four shelling plants, inoperative for periods of 2 to 16 years, were sampled for their insect fauna. The drug-store beetle; lesser mealworm; broad-horned fungus beetle; and two dermestids, Attagenus elongatulus and Attagenus megatoma, were collected. None of these insects, however, have been found in operating shelling plants.

Spider beetles, <u>Gibbium psylloides</u>, and the dermestid, <u>Anthrenus verbasci</u>, were routinely taken in samples from closed shelling plants but rarely are found in operating plants. Silverfish were the only live insects found in peanuts from the shelling plant closed for 16 years. <u>Trogoderma inclusum</u> larvae from a closed shelling plant at Oglethorpe, Ga., were heavily infected by the disease, <u>Mattesia trogodermae</u>. (SP 53-5)

# B. Biological and Physical Control

1. Pathogens. Many insects collected from peanut shelling plants were infected with disease organisms. Bacillus thuringiensis and a nucleopolyhedrosis virus were the dominant pathogens of almond moth larvae. A granulosis virus was the principal disease of Indian-meal moth larvae. A new species of Nosema was identified from corn sap beetle larvae collected from a peanut shelling plant. (SP 53-6)

# C. Improved Insecticidal Control

- 1. Protectants Intermediate Evaluation. Bayer 77488 was outstanding as a protectant for farmers stock peanuts for 1 year. Also, after 6 months, a combination of dichlorvos and malathion was still effective. The combination controlled initial infestations and protected the peanuts from insect invasion. (SP 53-1)
- 2. Space Treatments. Dichlorvos resin strips have protected untreated peanuts in small warehouses from moth infestation for 2 months in a test that is continuing. New strips will replace the old ones at 3 months. (SP 53-2)
- 3. <u>Peanut Fumigation Studies</u>. Residues in peanuts following fumigation with phosphine were less than 0.004 p.p.m. Also, there were no significant differences between fumigated and nonfumigated peanuts in organoleptic tests. (SP 53-8)

Publications - USDA and Cooperative Program
Thompson, John V., and Redlinger, Leonard M. 1968. Isolation of a nuclear polyhedrosis virus from the almond moth Cadra cautella
J. Invertebrate Path. 10(2):441-444. (SP-53)

#### AREA 8

### WOOL AND MOHAIR

Research Activity Code: 2-43-412-14-610

## USDA and Cooperative Program

Location of Intramural Work	: Scientist Man-Years F.Y. 1969 : Research Problem Area : : 408 : Total
Georgia	: 1.1 : 1.1 : 1.1
Total	: 1.1 : 1.1

No extramural support

## Progress - USDA and Cooperative Program

# A. Nontoxic Mothproofing

- Preliminary Evaluation of Protectants. Four organo-phosphorous compounds and a synthetic pyrethroid-type insecticide were the most promising of 29 compounds evaluated as potential mothproofers. Two of these compounds, Neopynamin and Ciba C-9491, are of special interest because of their performance and low mammalian toxicity. (SP 55-1)
- 2. <u>Improvement of Mothproofing Treatments</u>. Gardona applied to woolen cloth in practical home-type applications resulting in initial deposits ranging from 0.12 to 0.38% by weight still satisfactorily protected the cloth against black carpet beetle larval feeding after the treatments had aged 1 year.

Gardona was successfully applied to woolen cloth from Stoddard solvent and perchloroethylene under conditions simulating those of commercial drycleaning. The initial Gardona deposit on the cloth was found to be directly proportional to the Gardona concentration in the drycleaning solvents. There was no buildup of deposit after repeated drycleanings and retreatments. Steam pressing and dry heat had no effect on the deposit.

Bay compounds 75546 and 79845 were formulated as emulsifiable concentrates and applied at bath concentrations ranging from 0.05 to 0.20%. Cloths treated at all bath concentrations were satisfactorily protected against black carpet beetle larval feeding in precleansing tests. (SP 55-2)

3. Commercial Application of Quaternary Ammonium Compounds. Studies completed with Aliquat 336, an eight-carbon chain quaternary ammonium compound, include investigation by CSMA insect-feeding test of auxiliary treatments and automatic home washer applications.

Woolen cloth treated with Aliquat 336 at 0.5% by weight had increased wash resistance after treatment with Dow Corning XC-2-0510 silicone water repellent. Mykon SF polyethylene softener slightly increased the resistance of Aliquat 336 to drycleaning.

Woolen cloths treated with Aliquat 336 at 0.6% by weight in automatic washer applications were satisfactorily protected against both black carpet beetle and webbing clothes moth larval feeding in CSMA insect-feeding tests. (SP 13(C8898))

# Publications - USDA and Cooperative Program

- Anonymous. 1968. DDT replacement found as wool mothproofer. Oil, Paint, and Drug Reporter, 195(25):5. (SP-55)
- Bry, Roy E., McDonald, L. L., and Lang, Joe H. 1968. Allethrin as a protectant of woolen fabric against black carpet beetle larval damage. J. Econ. Entomol. 61(5):1194-1197. (SP-55)
- Bry, R. E., and Lang, J. H. 1968. Gardona as a protectant of a woolen fabric. Bul. Ent. Soc. Amer. 14(3):218. (SP-55)
- Happich, W. F., Windus, W., Naghski, J., and Bry, Roy E. 1968. Moth resistance of glutaraldehyde-stabilized wool. Textile Res. Jour. 38(9): 973-974. (Letter to the Editor) (SP-55)

#### AREA 11

#### TOBACCO

Research Activity Code: 1-23-408-14-608

## USDA and Cooperative Program

		Scientist Man-Years F.Y. 1969			
Location of Intramural Work	Research Problem Are	a : : Total			
Virginia	2	: 2			
Total	2	2			

No extramural support

# Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

- 1. Responses of Cigarette Beetle to Temperature Gradient: Cigarette beetle larvae exposed to a temperature gradient on a heat bar appear to prefer a temperature of  $82 \pm 4^{\circ}$  F. There was little larval movement in 24 hours at temperatures below  $68^{\circ}$  F. or above  $94^{\circ}$  F.; however, with longer exposures there was some larval movement toward warmer temperatures even from  $60^{\circ}$  F. (SP 16-1)
- 2. Mutation of the Tobacco Moth. Various degrees of redness were observed in the eye color of the  $F_2$  generation of the mutant tobacco moth. The moth was found not to be blind and is distinguishable in early larval instars from the wild type. Larvae with the gene(s) for the red eye color are white in early instars, whereas, they are pink in the wild type. (SP 16-8)
- 3. Cigarette Beetle Water Balance and Humidity Relationship. Cigarette beetle larvae developed on cornmeal fortified with yeast at 25°C without mortality at relative humidities of 73, 60, 53, and 43%. At 33% R.H., 65% mortality occurred; generally the larvae were in the 1st instar. At 13% R.H., all larvae died in 1st instar. At relative humidities of 43% or lower, larvae continued to molt.

At 73% and 93% R.H. and 25° C., partially dehydrated 3d and 4th instar larvae took up sufficient vapor from the atmosphere to gain approximately 10% in weight within a 24-hr. period. At 0% R.H. the larvae lost 10% in weight in approximately 72 hr. Vapor uptake was more pronounced in the 2d instar larvae than in the 3d and 4th.

Normal 3d instar larvae or larvae under negative water stress when offered a selection of 2 humidity levels, consistently selected the lower R.H. level. Even dehydrated 3d instar larvae preferred an R.H. level of 13% over one of 33%. (SP 72(A9795))

# B. Biological and Physical Control

- 1. Pathogens of Cigarette Beetle. A disease affecting cigarette beetle larvae has been identified as Bacillus sp. Studies on the mode of transmission of the spores show that the adult can carry the pathogen to clean media. Adults that emerged from treated media were allowed to oviposit in clean media. The disease was present in the progeny. (SP 16-7)
- 2. Attractants of Cigarette Beetle. Reliable isolation and purification procedures for the sex-attractants of the cigarette beetle have been established. Extraction by ether and sweep co-distillation procedures yielded the most reproducible and satisfactory results with respect to the final quantity as well as the purity of the attractants. The attractants have been eluted from GIC columns over a wide range of temperatures. Isolation and purification work is proceeding with a small portion of material that is quite volatile and with a greater portion of active material that is less volatile and more stable. (SP 71(A9785))

# C. <u>Improved Insecticidal Control</u>

- 1. Preliminary Insecticide Evaluation. Gardona® deposits originally of 100 mg/ft² after aging 24 months, continued to kill all exposed adult cigarette beetles. The same level of deposit of ENT-27386-4 (ethylmercaptophenylacetate S-ester with 0, 0 dimethyl phosphorodithioate) remained effective after 18 months. (SP 16-4)
- 2. <u>Vacuum Fumigation</u>. Vacuum fumigations of metal containerized units of tobacco were ineffective with acrylonitrile 34% plus carbon tetrachloride 66% at the rate of 5 pounds per 1,000 cubic feet. Larval survival in the tobacco at depths beyond 5 inches was high. (SP 16-5)

3. Atmospheric Fumigation. Five flue-cured tobacco warehouses ranging in size from 400,000 to 1.5 million cubic feet were fumigated for 4 days with phosphine at the rate of 20 aluminum phosphide tablets per 1,000 cubic feet. Peak concentrations of 300 to 500 p.p.m. occurring after 24 and 48 hours were sufficient to kill all insects at all depths in the tobacco hogsheads. Two days of aeration were required for the fumigant to leave the tobacco. (SP 16-6)

Respective exposures of 56, 48, 56, and 16 hours to phosphine concentrations of 90 to 100 p.p.m. were required for 100% mortality of unprotected cigarette beetle eggs, larvae, pupae, and adults. (SP 16-6)

# D. Insect-Resistant Packaging

1. Packaging Protectants. Liners, insecticides, and combinations of the two were not completely effective in protecting tobacco prized in boxes. The boxes of tobacco had been exposed to heavy infestation of the cigarette beetle for one year. Liners and insecticides tested were 1-mil polypropylene and polycarbonate and 4-mil polyethylene films, Bay 77488, Dursban®, lindane, and synergized pyrethrins. (SP 16-2)

# Publications - USDA and Cooperative Program

- Childs, Dana P. 1969. Phosphine Something new in fumigation. Tobacco 168(17:77. (SP-16)
- Childs, Dana P., Overby, James E., and Guy, Richard H. 1968. Laboratory evaluation of insecticides against the cigarette beetle, II. J. Econ. Entomol. 61(4):981-983. (SP-16)
- Childs, Dana P., Overby, James E., and Niffenegger, Dan. 1969. Phosphine fumigation of flue-cured tobacco warehouses for control of the cigarette beetle. Tobacco 168(21):20-25. (SP-16)
- Childs, Dana P., Overby, James E., and Watkins, Betty J. 1968. Low temperature effect on cigarette beetle infestation in tobacco hogsheads. J. Econ. Entomol. 61(4):992-996. (SP-16)
- Childs, Dana P., Overby, James E., and Watkins, Betty J. 1968. Vacuum fumigation of tobacco for control of the cigarette beetle. Tobacco Sci. 167(17):20-22. (SP-16)
- Childs, Dana P., Overby, James E., and Watkins, Betty J. 1968. Low temperature effect on cigarette beetle larvae. Bul. Ent. Soc. Amer. 14(3):212. (SP-16)
- Fletcher, Lowell W., Childs, Dana P., and Long, John S. 1969. An infestation of Trogoderma glabrum (Herbst) in cigarettes. U.S. Dept. Agr. Coop. Econ. Insect Rpt. 19(15)270. (SP-16)

#### AREA 13

#### INSECT CONTROL IN MARKETING CHANNELS - CROSS COMMODITY

The research reported in Area 13 is applicable to several commodities. For fiscal accounting purposes the level of input is somewhat arbitrarily allocated among three Research Problem Areas and the ten Research Activities of the Branch. The research program and the progress reports are not so organized.

## USDA and Cooperative Program

		:		Scie	ntist	Man	-Years	F.Y.	1969
Location of Intr	amural Work	:	Rese	arch	Probl	em	Area 1/	:	Total
		:	404	:	408	:	412	:	
		:		:		:		•	
Georgia		•	1.2	:	13.8	:	.8		15.8
California		:	•5		2.0	:	•5	:	3.0
		:		:		:		:	
		:		:	_	:		:	- 0 0
Total		:	1.7	:	15.8	:	1.3	:	18.8
		:		:		•		:	

No extramural support.

## Progress - USDA and Cooperative Program

# A. Biology, Ecology, Physiology, and Nutrition

1. Mitochondrial Metabolism in the Indian-Meal Moth. Under rearing conditions of 30°C., 75% R. H., and continuous light, Plodia interpunctella larvae reached a maximum average weight of 14 mg. 13 days after seeding the cultures with newly laid eggs. The average pupal weight was 13.5 mg. A slight weight loss occurred during the 6-day pupal period. Peak adult emergence occurred on day 21. Initial male weight was 8 mg., and the weight decreased about 1 mg. with age. Initial female weight was 12 mg., and weight decreased 9 mg. within a week. Larvae weighing <6 mg. died, and those weighing >8 mg. pupated within 3 days after starvation was initiated.

Electron microscopy of the mitochondrial fraction isolated from <u>Plodia</u> larvae showed virtually no contamination by other subcellular organelles. Comparison of isolated mitochondria with those found <u>in situ</u> indicated structural integrity was largely retained.

Aspartate-pyruvate transaminase and lactic dehydrogenase activities increased during the period when <u>Plodia</u> larvae accumulated lipids. Other transaminase activities located in the cytosol fraction and which are most intimately associated with mitochondrial metabolism did not significantly change during larval growth. These data suggest that larval accumulation of lipids has little influence on protein catabolism. (SP-65)

Photoperiod Studies With the Indian-Meal Moth. Continuous light had 2. several effects on the reproductive behavior of Plodia interpunctella. Adults emerging from cultures reared in continuous light were slower in mating. Since oviposition is dependent upon a stimulus for mating, the ovipositional period was therefore prolonged. The total egg production per female in continuous light was significantly lower than in 12 hr.:12 hr. light-dark conditions when females were mated with males reared in continuous light. A comparison of spermathecal contents in females mated to males from continuous light and light-dark cycles showed that most of the former contained few spermatozoa in the spermathecae. Therefore, oviposition in P. interpunctella appears to be triggered by the quantity of spermatozoa in the spermathecae of the female rather than by the mechanical or physical stimulus of the copulation. In order to assess effective mating and oviposition potential in P. interpunctella, it is necessary that spermathecal contents be examined rather than the spermatophore.

Analysis of the hourly samples of emerging adults revealed several aspects of the rhythmic development of P. interpunctella. Under alternating 12-hr light and dark cycles, the mean time of each peak was approximately 20-26 hours apart. The mean peaks occurred approximately at the same time of each 24-hr. cycle. The rhythm synchronized the emergence of the population and 95% of each daily complement emerged within an 8-hr. period. In contrast, emergence in continuous light lacked this synchrony and occurred without interruption. (SP-66)

3. <u>Humidity and Insect Behavior</u>. Fourth instar larvae and adults of Oryzaephilus surinamensis were observed in circular two-way choice chambers. The opposing halves of the chambers differed in percentage of relative humidity, intensity of light, or degree of tactile stimulation, or in two of these factors. The humidity response in the larvae appears to be brought about by an oriented response (taxis). The larvae show a strong preference for areas in which they are provided increased tactile stimulation. The response to tactile stimuli is dominant over the humidity reaction.

Adult males showed a hygronegative response to gradients between 100-60 and 80-40% R.H. but were indifferent to 60-20 and 40-0% R.H. Adult males desiccated for 48 hours showed a slight hygropositive response in a 100-60% R.H. gradient.

Adults showed a strong photonegative response which interacted with the humidity reaction. Behavioral and morphological investigations indicated that the hygroreceptors are located on the antennal club (terminal three segments) in adult males. (SP-67)

4. Low Temperature Adaptation in Stored-Product Insects. Adults of Tribolium confusum were acclimated at high (30°C.) and low (15°C.) temperatures for use in a study of dispersal through a relatively large flour bulk. The dispersal chambers (cubic foot boxes divisible with sliding partitions into 64 smaller compartments) were filled with a 1.1 mixture of cornmeal and white flour and preconditioned for 24-48 hours at 5, 10, 15, 25, and 30°C. Insects were then placed at the centers of the chambers. The chambers were partitioned, and after 1, 3, 5, 7, and 11 days the precise locations of the insects determined. The results show that thermal acclimation has a persistent influence (at least 11 days) on the distribution of the beetles in flour stored at moderately low temperatures. At higher storage temperatures the effect of acclimation is briefer; the higher storage temperature overrides the greater activity of the cold-acclimated animals and equalizes final distributions.

In the research on the rate of acclimation at organismal (locomotory), metabolic (O<sub>2</sub>), and enzymic (ATP-ases of muscle) levels, the following levels of study were conducted: <u>T. confusum</u> at organismal, metabolic, and enzymic; <u>Trogoderma parabile at organismal and enzymic; Sitophilus oryzae at organismal; Tenebrio molitor at metabolic; and <u>Musca domestica at organismal</u>, enzymic, and metabolic. The house fly was inserted to provide data for an insect radically different in habitat requirements and behavioral and metabolic demands. Results of these studies are now being prepared for publication. (SP-17(G8071))</u>

5. Bionomics of Trogoderma. Characters suitable for sexing pupae with 100% accuracy were found in the adult secondary sexual characters of the antennae and last abdominal segment. A manuscript is being prepared on this subject for publication.

A total of 376 samples were diagnosed for disease infection. Mattesia trogodermae continued to be the most common pathogen observed and was found in 74% of the field samples examined. Eugregarines were found in 13%, a microsporidian and the coccidian, Adelina tribolii, in less than 1%. Description of the eugregarine has been complicated by the possibility of two species being involved.

Studies on the influence of constant temperature and humidity on growth and development of  $\underline{\text{Trogoderma}}$  glabrum,  $\underline{\text{T. parabile}}$ , and  $\underline{\text{T. inclusum}}$  were nearly completed.

The massive amount of data from field collections was tabulated and preparation was started on the first of a series of reports on the distribution and abundance of <u>Trogoderma</u> and associated species of stored-product insects. The first report being prepared is concerned with the method of obtaining sample populations.

Food studies, involving more than 20 kinds of foods and using both mature larvae and adults in parallel tests, were made with  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{glabrum}}$ ,  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{inclusum}}$ , and  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{parabile}}$ . Tests with  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{simplex}}$  were made with mature larvae only. Samples were prepared to complete studies with  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{simplex}}$  adults and start studies with  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{grassmani}}$  and  $\underline{\mathtt{T}}$ .  $\underline{\mathtt{sternale}}$ .  $\underline{\mathtt{(SP-18(G8073))}}$ 

6. <u>Nutrition of Almond Moth</u>. Tests were made to determine the need for individual mineral salts and their optimal concentrations in the larval diet for the almond moth. A new salt mixture was formulated to replace the salt mixture (M-D No. 185) which was used previously in the vitamin study diet. Results indicate that the new salt mixture provides the necessary minerals and that insect growth was satisfactory.

Preliminary tests were also made to determine whether a mixture of amino acids can replace casein as a source of protein in the original diet. Growth on these test diets appeared to be as good as that on the ones containing casein. (SP-23(A8421))

7. Use of Food by Stored-Product Insects. An enzymatic spectrophotometric method for the determination of uric acid was modified and applied to the work on this project. As a result, it is possible to determine accurately the uric acid content of insect feces. Details of this method have been decribed in a manuscript submitted for publication.

Utilizing this enzymatic method, it is now possible to estimate the consumption of food by measuring the fecal uric acid produced by insects over a given time. The method consists of separating a small sample of feces and computing the uric acid content. A mixture of food and fecal material from an insect colony is then homogenized and an aliquot analyzed for uric acid. Since the food material contains no uric acid, the weight of feces present can be calculated by dividing the total weight of uric acid in the mixture by the weight of uric acid per known weight of feces. The method gave results almost identical to those obtained by manual separation of food and feces with a subsequent determination of the uric acid content in the separated feces. (SP-24(A9031))

8. Microorganisms from Alimentary Canals of Beetles. Pathogenicity studies of Escherichia coli serotypes to chicken embryos were continued. E. coli serotypes recovered from the lesser mealworm, Alphitobius

diaperinus, were compared with similar serotypes isolated from poultry. E. coli recovered from the insects were generally less pathogenic to the embryos. Subsequent studies with 19 E. coli isolates from the lesser mealworm indicated the differences in pathogenicity resulted primarily from host specificity.

Gut transmission and retention studies involving the mycotoxin F-2 were conducted on the confused flour beetle and the lesser mealworm. The mycotoxin was retained and remained viable in the insect's gut. There was little reduction in quantity of F-2 within the insects unless they were starved, which indicated that some of the F-2 was passed with the feces.

The effect of F-2, with and without yeast as a diet additive, on the fecundity and fertility of the confused flour beetle, was investigated.

Adult lesser mealworms were collected from chicken and turkey brooder houses to be examined for the presence of <u>Salmonella</u> spp. Samples of mealworm dust and litter were also analyzed to determine possible relationships between the insects and their environment. (SP-26(A9357))

9. Amino Acids and Vitamins in Physiology of Insects. Research on the project involved the investigation of amino acid and vitamin requirements and deficiency effects on the physiology of larval, pupal, and adult life stages of the test insects. Results to July, 1969, have been qualitative and relate only to Oryzaephilus surinamensis. The recovery of methionine from adults and alanine from pupae as well as the determination of the essential B vitamins is significant, and the knowledge possibly will offer some opportunity to effect insect population reductions. A more critical evaluation of this potential must await the results of rearing experiments presently underway but not completed · (SP-54(A7-MQ-18))

# B. Biological and Physical Control

1. Controlled Atmospheres. Humidity has been found to be an important adjunct in the use of modified atmospheres for insect control. One-day exposures to atmospheres containing approximately 2.6% of oxygen and 97.4% of nitrogen at 68% and 33% R.H. produced 5% or less and over 95% mortality, respectively, in the red and confused flour beetles. A 3-day exposure to an atmosphere composed of 2.6% nitrogen and 97.4% oxygen at 30% and 56% R.H. produced 100% and 70% mortality, respectively, in the saw-toothed grain beetle. Only a 2% difference in mortality was found when male and female red flour beetles were exposed separately to a ternary atmosphere containing 47% carbon dioxide and 12% oxygen. Red flour beetles cease reproduction at a concentration of carbon dioxide between 16% and 22% when the oxygen concentration is maintained near that of the normal atmosphere.

Wheat infested with the lesser grain borer adults for 1 week was held for 1, 2, 3, 4, or 5 weeks and then exposed for 1, 2, 3, or 4 days to various concentrations of atmospheric gases. Concentrations of 100% nitrogen or nitrogen with a small amount of oxygen were not as effective in reducing emergence as were ternary mixtures containing 46% or more of carbon dioxide. The only mixtures effective after 5 weeks of infestation were ternary atmospheres having 59% or more of carbon dioxide. (SP-56)

- 2. Light. Photoresponse tests of black carpet beetles showed behavior patterns that agree with spectral sensitivities determined by electrophysiological studies. Maximum behavioral response occurred at wavelength peaks derived from electroretinograms. However, in color choice tests intensity-dependent reversals in behavior were noted. With equal intensities of green and UV at the 0.02 mw./cm.² level, adult beetles showed distinct preference for UV. This could be reversed by increasing the intensity of green about tenfold. Low intensity tests at the 2.5 nanowatt/ cm.² level showed preference for green over UV at equal intensities. Photonegative larvae moved from green to UV at this low intensity level. Further tests are underway to clarify this behavior. Single flash electroretinograms of this beetle show that functional maturation parallels morphological maturation in this species. The eye is not functionally mature until about 9 days after emergence. Study of the compound eye of a "blind" laboratory strain of Tribolium castaneum shows retention of some degree of function at UV, suggesting a possible separate receptor system for UV in this eye. (SP-57)
- 3. Gamma Radiation. Tests have been conducted in which dried fruits and nuts infested with eggs, young larvae, or adults of the saw-toothed grain beetle and the Indian-meal moth were irradiated. Complete control of all three life stages of both species was achieved by a 40-krad treatment.

A test was terminated after 14 months in which white wheat flour was infested with eggs and young larvae of the almond moth and the Indian-meal moth and with all life stages of the red and confused flour beetles and the saw-toothed grain beetle. One-half of the flour was untreated, and the other half was irradiated at 30-50 krad. Sampling was conducted periodically during the test period. No live insects were found in the treated flour after 3 months.

Nine 500-bushel bins of infested wheat, irradiated at 0, 20-30, or 40-50 krad in October, are being periodically sampled. After 6 months' storage, the 0 treatment had an average of 23.3 adult and 100.6 immature insects, the 20-30-krad treatment had an average of 0.4 adult and 5.6 immature insects, and the 40-50-krad treatment had an average of 0.1 adult and 2.3 immature insects per 1,000-gram sample. (SP-58)

There was no significant difference in total population development between the control and populations started with male Acarus siro irradiated with 5 krad. A significant difference was observed between populations started with males irradiated at 5 and 15 krad. Irradiating males with 15 krad was effective in reducing the population development 14-fold. There were no significant differences in populations initiated with males irradiated with 15, 25, and 35 krad.

A squash technique was employed to examine chromosomes from gonads of male A. siro irradiated with 0, 1, 5, 10, 15, 25, and 50 krad. Chromosome stickiness was observed in gonads irradiated with 5, 10, 15, and 25 krad. No meiotic figures were observed for 7 days in mites irradiated with 50 krad. An apparent increase in the number of midmeiotic figures (metaphase and anaphase) was observed in gonads irradiated with 1 and 5 krad. A delay in the appearance of normal meiotic figures increased with an increase in radiation dosage.

Post-irradiation respiration studies of adult female A. siro indicate no difference between 0, 5, 10, and 15 krad. A variable difference occurred at 25 krad.

Gonads from male and female A. siro have been cultured in vitro. Aceto-orcein-stained squashes of male gonads have revealed a variable chromosome number of 2N = 16 and 2N = 17. (SP-30(A9408))

- Production of Sound by Insects. Feeding sounds were detected with 4. rice weevils in wheat and corn, lesser grain borers in wheat, Angoumois grain moths in corn, and cowpea weevils in cowpeas and black-eyed peas. In timed listening tests with single kernels of wheat infested with rice weevil, there was considerable variability from test to test. Parallel simultaneous monitoring of many kernels appears to be the best method. An adhesive-coated plastic sheet has been adopted as a means of sample gathering and conducting the sound to the transducer. Continuous monitoring throughout the life cycle of rice weevil in wheat has not revealed any diurnal variations in feeding or activity; however, these data are still being gathered. Preliminary tests using microwaves to detect internal infestation of soft red winter wheat showed that small but measurable amounts of energy are absorbed in the microwave region. Immature rice weevil infestations averaging 1, 2, 3, and 4 weeks of age were detected at 10.7 and 11.2 GHz at 0.04 and 0.02 µw. power levels. (SP-63)
- 5. <u>Biophysical Research</u>. A technique for the definite identification of hygroreceptive sensillae was developed. This will permit the correlation of structure and function in these organs.

Experiments aimed at identifying a pheromone-receptor protein in the male Plodia antenna were prepared.

The morphology and function of the eye of the red flour beetle have been studied, and publications are in preparation.

The morphology of the midgut of the Indian-meal moth has been thoroughly defined, and its changes in response to starvation are established. Studies of lipid uptake and radiation response were initiated. (SP-64)

6. Grain Chilling. Five virgin adult male and five virgin adult female red flour beetles were confined in each of several jars containing media and exposed to temperatures of 40°, 50°, 60°, 70°, and 80° F. for 1, 2, 3, and 4 weeks. Immediately after exposure, the adults were removed from the media and mortality counts were made. At 40° F., no mortality occurred after a 1-week exposure but all adults were dead after a 4-week exposure. After a 4-week exposure at 50° F., 30% of the adults were dead. The live adults were removed from all tests after 4 weeks, and the media was then held at 80° F. for another 7 weeks. No F1 insects were found in the media in which the adults were exposed at 40°, 50°, and 60° F.

A similar test was run with confused flour beetles. At  $40^{\circ}$  F., 30% mortality was found after a 1-week exposure and all adults were dead after a 3-week exposure. No  $F_1$  insects were found in the media in which the adults were exposed at  $40^{\circ}$ ,  $50^{\circ}$ , and  $60^{\circ}$  F. after it was held another 7 weeks at  $80^{\circ}$  F.

Two 500-bushel metal bins were erected, and one was insulated with 2-inch styrofoam and the other with 3-inch styrofoam. Plenum chambers for distribution of the cool air from the grain-chilling unit have been installed in these bins. The grain chiller has been equipped with a watthour meter to measure power consumption. (SP-76)

7. Chemical Constituents of Foods to Control Insects. Damage to five southeastern soybean varieties by cowpea weevils after 120 days in the laboratory ranged from 235 to 7 per 500 gram sample revealed their susceptibility as follows: Coker 102>Coker 208>Bragg>Hampton>Davis. The quantitative deviations of fat-oil, saponin, and genistin in Davis, Coker 208, and Hampton varieties of soybean were too small to be the decisive factors influencing the degree of infestation by cowpea weevils.

A slight delay in cowpea weevil development in artificial beans occurred when the amount of soybean, lima bean, and pinto bean flours added to black-eyed pea flour was increased. Dry lemon or tangerine peel powders, or commercial saponin, prevented cowpea weevil development in the artificial beans.

Wheat was dusted with various soybean components as a protectant from rice weevils. Ether-defatted soybean and soybean saponin were highly toxic; soybean flour was less toxic; and defatted desaponin soybean was nontoxic.

Fresh citrus peel oils as a surface treatment to black-eyed peas provided protection from cowpea weevil oviposition in the following order: tangerine, kumquat, lemon (good); grapefruit, lime (moderate); orange, tangelo, temple orange (poor). The topical application of the oils on cowpea weevils indicated toxicity in the order: lemon, grapefruit, lime (high); kumquat, tangerine (medium); orange, tangelo, temple orange (low). (SP-77)

8. Pathogens. Thirty accessions of about 2,500 larvae and adults of stored-product insects were received for diagnosis. Many of these specimens harbored viral, bacterial, and protozoan pathogens. Eleven previously unknown pathogens were isolated from this material, making a total of about 45 pathogenic microorganisms that have now been found in various stored-product insects.

Tests indicated that Microsporidia continue to exert a biological control which extends beyond the generation that is initially infected. Female Indian-meal moths, Plodia interpunctella with even inapparent infections of Nosema plodiae laid significantly fewer fertile eggs than did healthy females when both were paired with healthy males; furthermore, there was a decrease in the number of fertile eggs laid by healthy females paired with diseased males. Nineteen percent of the progenies of diseased females acquired infections transovarially. Five percent of the venereally infected females transmitted Nosema infections to their progenies.

A previously unreported granulosis virus was isolated from larvae of the almond moth, <u>Cadra cautella</u>. The virus is highly pathogenic and cross-infective to the <u>Indian-meal</u> moth. Preliminary electron microscope examination of cross-infected larvae showed aberrant encapsulation of the virus particles. (SP-78)

9. Effect of Sounds on Insects. New insect-mounting or tethering techniques were devised to permit separating airborne sounds from solid-borne substrate-conducted vibrations. Tests with Tribolium confusum, Sitophilus oryzae, Attagenus megatoma, and Trogoderma inclusum indicate duplex reception of sounds. All species gave positive responses when mounted by fastening the pronotum or elytra to conical wax blocks attached to thin applicator sticks. By alternate method, insects were mounted to ends of cotton thread soaked in a low melting point mixture of paraffin and mineral oil.

In general, midrange sensitivity was estimated at 40-50 db, falling to 100-110 db (equipment limits) at range extremes.

In block-mounted  $\underline{S}$  oryzae, 80% responded to sounds between 20 and 400 Hz. With thread mounting, only 20% responded to sound between 20 and 150 Hz.; 80% gave no response. Detergent water solutions applied to the bodies to load or change resonance of hairs gave inconclusive results.

Block-mounted A. megatoma responded between 20 and 250 Hz. Detergent solutions stopped these responses as did thread mounting.

Block-mounted <u>T. inclusum</u> responded at 90° F., but not at 70° F. Fifty percent of the thread-mounted insects responded from 1,600 to 3,100 Hz.

Block-mounted  $\underline{\mathbf{T}}$ . confusum responded from 35 to 150 Hz, with only 5% negative results. Thread mounting extended the upper range to about 275 Hz. (SP-21(G9196))

Sound perception in <u>Plodia interpunctella</u> was confirmed indirectly by observation of behavioral responses to sounds produced by loud-speakers, bells, and whistles. These sounds caused cessation of wing motion in moths mounted on a flight mill. Free-flying moths either dropped to the ground or changed flight direction with the occurrence of sound stimuli. There were indications of sex-related differences in frequency sensitivity to sounds in the range of 2 to 40 kHz.

Morphological studies showed the presence of tympanic organs on the first abdominal segment just posterior to the constriction between the abdomen and the thorax. Unlike noctuid moths, the tympanic cavity of <u>Plodia</u> consists of only one chamber. The tympanic nerves arise from the first abdominal ganglion. (SP-29(A9407))

10. Sex Attractant of Indian-Meal Moth. The sex attractant in Plodia interpunctella has been isolated and purified for identification. Approximately 200  $\mu$ g. have been submitted for identification. Synthesis of the compound will be undertaken following identification.

In laboratory bioassays, the isolated compound was highly attractive. A significant number of males responded to a concentration of  $10^{-12}~\mu g$ ., indicating that this pheromone is as strong as any known pheromone from insects. Bioassays indicate neither the crude nor the GIC purified attractant was significantly more active than the other. GIC studies indicate that there is only one compound in the sex pheromone. It has been concluded that contaminants in the crude extracts are not masking the attractant's activity and that there are no synergist-like compounds supplementing its activity. These results also indicate that crude extracts are giving a true picture of the differing pheromone concentrations found in moths of different ages.

In a number of trapping surveys using virgin females as lures, male <u>Plodia</u> were caught from a variety of habitats; i.e., peach and pecan orchards, residential and farm areas, and storage areas. This indicates that the species is common to many areas, and explains how a commodity can be infested in the field before being placed in storage. (SP-31(A9409)

11. Neuroendocrine System in Sitophilus. The first phase of the study nearing completion is a generalized description of the morphology of the neuroendocrine and reproductive systems and the determination of the total body chemical composition. From these data, relationships will be formulated as they relate to the possible influences of the neuroendocrine system on reproduction and its nutritional adjustments.

There appears to be a correlation between reproductive organ development of the virgin and mated females and the pattern of the neuroendocrine system development, but it is still too early to conclude that the correlation is indeed a relationship. The metabolic data will be analyzed in a similar fashion with the pattern of neuroendocrine development in order to obtain any possible correlations that might exist between these two systems. (SP-32(A9413))

12. Parasites and Predators of Indian-Meal Moth. The research activities on this project have provided sufficient material for the preparation of two graduate student theses. The host-parasite relationships between the Indian-meal moth and the parasites, Bracon habetor and Melichares tarsalis have been investigated. It was found that under replicated experimental conditions an Indian-meal moth population of 200 adults was progressively reduced by increasing the number of introduced Bracon up to 12 pairs. Introduction of 24 pairs of Bracon resulted in no control over a 4-week period. Tests are in progress to determine the best time for parasite introduction with respect to the developing larvae. Stock colonies of moths also are being reinoculated with moths at definite intervals to determine the proper timing necessary to maintain a population of the parasite.

Information has been collected on the life history and feeding habits of <u>Melichares tarsalis</u>. The results have been analyzed and submitted for <u>publication</u>. (SP-33(A9421))

13. Quinones to Control <u>Tribolium</u>. Research on the effects of natural and artificial quinone conditioning of food medium on the life stages of <u>Tribolium confusum</u> was completed. Subsequent investigations of the <u>Indian-meal moth</u>, the cigarette beetle, the saw-toothed grain beetle, and long-headed flour beetle revealed that the saw-toothed grain beetle and the long-headed flour beetle were quinone-secreting insects.

The long-headed flour beetle was selected as a test organism to further evaluate the biological effects of natural and artificial quinone conditioning. Concentrations of the quinone materials in the test treatments

were set at 250, 500, 1,000, and 2,000 p.p.m. (by weight) for the quinones, p-benzoquinone and methyl p-benzoquinone. Initial interpretation of the raw data indicates that the egg viability of all stages is negligibly affected by both natural and artificial quinone conditioning within the concentration levels used and the fecundity is unaffected by synthetic quinones; however it is significantly reduced by natural conditioning. (SP-36(A9708))

14. Masking Sex Attractants. A synthesized sex attractant of the black carpet beetle was tested on males of Attagenus megatoma. The males were easily attracted, and exhibited a series of primary responses characteristic of normal mating behavior. The compound, trans-3-cis-5-tetradecadienoic acid, elicited responses in the males which were identical to those exhibited by them in the presence of virgin females.

Several related compounds were tested for activity as sex attractants and two isomers of tetradecadienoic acid, <u>cis-3-trans-5-tetradecadienoic</u> acid and <u>trans-3-trans-5</u> tetradecadienoic acid, showed potential as masking agents for the natural sex attractant in the black carpet beetle. (SP-75(A9805))

15. Physiological Control. Research emphasis on this project has been shifted temporarily from the use of antimetabolites in insect control to the investigation of insect nutrition. Evidence has been presented that shows that the effectiveness of antimetabolites can affect insect sterility, longevity, and reproduction. However, the materials examined are in most instances highly toxic to mankind and, therefore, do not offer promise in insect control.

Nutritional investigations have indicated some significant areas of research in the use of rapid oxidants as dietary antagonists of essential amino acids. One dietary observation of interest is that overdoses of biotin, a B-vitamin, in the diet can cause sterilization in the adult hide beetle. (SP-40(AlO-MQ-4))

- 16. Effect of Vacuum on Insects. In a 10-mm. Hg.vacuum at 80° F. and 40% R.H., IT95 values for Indian-meal moths are 429.9, 99.0, 271.5, and 32.2 min. for eggs, larvae, pupae, and adults, respectively. (SP-61)
- C. Mode of Insecticide Action and Development of Resistance
- 1. Oxidative Detoxification of Enzymes. Heptachlor epoxide was the principal metabolite produced by different stages, ages, sexes, and strains of Plodia interpunctella and Trogoderma parabile when they were treated with acetone solutions of heptachlor. Epoxidative conversion values were calculated from the total heptachlor and heptachlor epoxide extracted from the insects by hexane.

Time-course studies with adults of the two species showed a more rapid epoxidative conversion of heptachlor by <u>Trogoderma</u> (47%) than by <u>Plodia</u> (27%) 10 hours after treatment. At 24 hours this difference was less significant, being 56% for <u>Trogoderma</u> and 46% for <u>Plodia</u>.

Plodia and Trogoderma metabolized heptachlor to heptachlor epoxide at about the same rate. Larvae of these two species were found to absorb heptachlor as rapidly as their respective adult stages. A significant difference in epoxidative ability was discovered between 10- to 12-day-old Trogoderma adults. Conversion of heptachlor to heptachlor epoxide was  $48 \pm 4\%$  for males and  $65 \pm 6\%$  for females. No significant differences were found between 3-day-old Trogoderma adults, the values being  $51 \pm 9\%$  for males and  $55 \pm 5\%$  for females.

Cultures of Trogoderma obtained from two locations, Savannah and Fresno, produced adults with different epoxidative capacities. Ten hours after topical application of heptachlor, adults from the Savannah cultures converted 55  $\pm$  7% of the heptachlor to heptachlor epoxide and those from the Fresno cultures converted 65  $\pm$  7% of the heptachlor to heptachlor epoxide. (MQ1-43(GR))

2. Resistance to Insecticides. Indian-meal moth larvae from corn in Kansas were found so resistant that 50% mortalities could not be obtained with 1  $\mu$ l. of 95% malathion. These larvae were only slightly resistant to Bay 77488, dichlorvos, and Dursban. Indian-meal moth larvae from stored peanuts in Georgia showed high resistance to malathion. Gardona and carbaryl synergized with Hoffman-LaRoche RO 5-8019 indicated promise against the resistant larvae.

Resistance in a flour beetle strain from peanuts in Georgia was 50.9-fold to malathion and 9.6-fold to DDT. (SP-61)

3. DDT Resistance in Khapra Beetle. Research on this project has increased our knowledge of the sterol requirements of Trogoderma granarium. The study included an investigation of the different life stages (larva, pupa, and adult) and presented some insight into sterol metabolism and into the amounts of lipids present and their breakdown into neutral lipids and phospholipids.

The finding that phosphatidyl ethanolamine was the main phospholipid in the larval and pupal stages is significant. Since this condition is in contrast to our knowledge of phospholipids in other coleopterans and mammals, it warrants investigation as a manipulative factor in host-plant breeding for possible insect pest population reduction. (SP-2(A7-MQ-25))

# D. Improved Insecticidal Control

- 1. <u>Preliminary Evaluation</u>. Of 60 new insecticides tested, 23 were promising for followup tests. LD values were determined for two compounds against two insect species.
- Of 45 compounds tested, 12 showed greater repellency than did the standard.

Dichlorvos spray deposits on wheat at 4.6 p.p.m. killed adult white-fringed beetles in 24 hours but not eggs. Most residues were removed with the bran and shorts in milling. Flour baking quality was unaffected.

Gardona, Bay 77488, and Ciba 9491 are promising as residual treatments on wood surfaces. (SP-61)

2. Practical Control Problems. A 30-lb. lot of dichlorvos pellets provided 16 vapor applications suitable for treating a 100,000 cu.-ft.-warehouse.

A 72-hr. fumigation of food items under plastic films with 100 aluminum phosphide pellets per 1,000 cu. ft. controlled insects. (SP-61)

# E. Insect-Resistant Packaging

l. Residue Barriers. Ethylene vinyl acetate resin in wax barrier coatings or in synergized pyrethrins-wax coatings on kraft paper reduced the migration of piperonyl butoxide into flour. A highly oriented laminated polyethylene film also appears to be a good barrier to insecticide migration. This material also appears to be somewhat resistant to insect penetration. Laboratory tests indicated that fat contents of 4% or less in flour did not affect the migration of piperonyl butoxide. However, 8% fat had a significant influence on migration of piperonyl butoxide. Creping saran-coated kraft did not impair insecticide barrier qualities; however, creped EVA-wax coated kraft was more effective than was similarly treated flat kraft. A large-scale, long-term test has been started to evaluate several barrier systems for their effectiveness in preventing the migration of piperonyl butoxide. (SP-59)

Experimental 25-lb. bags were delivered under terms of a contract for large-scale, long-term storage studies of insecticide barrier systems found effective in laboratory tests. The 20 barrier systems to be evaluated include kraft paper with saran coatings over styrene butadiene and polyethylene, polyvinyl alcohol-coated kraft, and glassine paper and greaseproof paper in bags having outer plies treated with either wax or clay containing synergized pyrethrins. The barrier systems are being tested with flour, cornmeal, and CSM (cornmeal-soybean meal-milk), which have fat contents ranging from less than 1 to almost 6%. (SP-10(C8174))

2. Insect-Resistant Cotton Bags. A shipping test to the Philippines was conducted to determine the resistance to infestation of two insect-resistant treated (IRT) cotton bags. One bag had an IRT kraft/cotton laminated outer ply, and the other had an outer ply of IRT cotton. Both bags had waxed, creped kraft liners and tape-over-stitch (TOS) bottom and top closures. Standard untreated cotton bags and IRT kraft bags were also included in the test. With good closures, none of the IRT cotton bags or IRT kraft bags were infested when they were unloaded in Manila about 2 months after they were filled. All of the untreated cotton bags contained insects. After 6 months' storage at two locations in the Philippines, nearly all of the IRT kraft/cotton laminated bags were free of insects. Nearly all IRT cotton bags were infested but with very few insects. The level of protection provided by the IRT kraft bags was intermediate between that of IRT kraft/cotton and IRT cotton bags.

An economical method was developed to treat cotton with synergized pyrethrins at the mill. The warp yarns were treated on the slasher before the fabric was woven. (SP-60)

## F. Fate and Effect of Residues

- 1. Analyses Made. During the past year 5,121 analyses were conducted in support of the various research programs within the Branch. The breakdown of analyses was as follows: 1,642 dichlorvos, 1,557 piperonyl butoxide, 1,182 malathion, 224 Gardona, 109 Bay 77488, 100 bromide, 100 chloride, 88 phosphine, 49 diazinon, 27 wax, 13 DDT, 11 fat, 5 lindane, 5 Dursban, 4 Accothion, 3 methoxychlor, and 2 pyrethrins. (SP-62)
- 2. Method Development. Methods were developed for residue analyses of Gardona and Bay 77488 applications on inshell peanuts, wool cloth, and plywood sheeting. (SP-62)

# G. Toxicology

1. Action of Fumigants on Animals. Research on this project has been preliminary in nature. An interesting finding has resulted from one initial test. This experiment has shown that fumigant residues from carbon tetrachloride, chloroform, trichlorethylene, and carbon disulfide disappear more rapidly from wheat, corn, barley, and sorghum at 17°C. than at 30°C. (SP-70(AlO-MQ-8))

# Publications - USDA and Cooperative Program

# Biology, Ecology, Physiology, and Nutrition

- Anderson, Richard L., and Mutchmor, John A. 1968. Rate of temperature acclimation in the confused flour beetle. Bul. Ent. Soc. Amer. 14(3):205. (SP-17)
- de las Casas, Ernesto., Pomeroy, B. S., and Harein, Phillip K. 1968.
  Infection and quantitative recovery of Salmonella typhimurium and
  Escherichia coli from within the lesser mealworm, Alphitobius
  diaperinus (Panzer). Poultry Sci. 47(6):1871-1875. (SP-26(A9357))
- Golebiowska, Zofia. 1968. Development and fertility of the Mediterranean flour moth Anagasta (Ephestia) kuhniella Zeller (Lepidoptera, Pyralidae) in differently coloured vessels. Ekologia polska Series A, XVI(23): 485-504. (E21-AMS-4)
- Harein, Phillip K., and de las Casas, Ernesto. 1968. Bacteria from granary weevils collected from laboratory colonies and field infestations. Jour. Econ. Ent. 61(6):1719-1720. (SP-26(A9357))
- Lum, P. T. M., and Flaherty, B. R. 1968. Effect of mating with Indianmeal moth males from constant light or light-dark cycles on oviposition. Bul. Ent. Soc. Amer. 14(3):210. (SP-66)
- Waites, Robert E., and Gothilf, Shmuel. 1969. Nutrition of the almond moth. I. Analysis and improvements of the experimental diet. Jour. Econ. Ent. 62(2):301-305. (SP-23)
- Yurkiewicz, William J., and Oelsner, J. 1969. Neutral lipid metabolism during embryonic development of the Indian-meal moth, Plodia interpunctella (Hubner). Compar. Biochem. Physiol. 28(2):955-959. (SP-65)

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- Brady, U. Eugene. 1969. Response of male <u>Sitotroga</u> to the sex pheromones of <u>Sitotroga</u> and <u>Pectinophora</u> females and to propylure and deet. <u>Deet analysis</u> in <u>Sitotroga</u> females. <u>Jour. Ga. Ent. Soc. 4(1):11-14</u> (SP-31)

- Brady, U. Eugene and Smithwick, Eleanor B. 1968. Production and release of sex attractant by the female Indian-meal moth, Plodia interpunctella Ann. Ent. Soc. Amer. 61(5):1260-1265. (SP-31)
- Brown, Gary A. 1968. Some effects of irradiated males on population development of <u>Acarus siro</u>. Bul. Ent. Soc. Amer. 14(3):210. (SP-30(A9408))
- Brown, Gary A. 1968. Some effects of irradiated males on population development of Acarus siro L. (Acarina: Acaridae) M. S. thesis, Univ. Ga. (SP-30(A9408))
- Butler, Linda, Roppel, R., and Zeigler, J. 1968. Age dependent changes in the eye of the black carpet beetle; an electron microscope study. Bul. Ent. Soc. Amer. 14(3):205. (SP-57 and SP-64)
- Cogburn, R. R., and Mahany, P. G. 1968. Gamma radiation effect of malathion on wheat and kraft paper. Bul. Ent. Soc. Amer. 14(3):218. (SP-58)
- Cohick, Albert Doyle, Jr. 1968. The effect of natural and artificial quinone conditioning of food media on life stages of <u>Tribolium</u> confusum. Ph. D. thesis. (SP-36)
- Continé, Yvette M. 1968. Effects of gamma radiation on the karyotype of in vitro cells of the grain mite. Bul. Ent. Soc. Amer. 14(3):210. (SP-30)
- Davis, R., and Brown, S. W. 1968. Life tables and intrinsic rates of natural increase for the grain mite. Bul. Ent. Soc. Amer. 14(3):210. (SP-30)
- De, R. K. 1968. Attractants for the rice weevil and the cowpea weevil in cereal and legume seeds. Bul. Ent. Soc. Amer. 14(3):209. (SP-19)
- Hemenway, Robert C. 1968. Respiration in the grain mite preceeding and following treatment with gamma radiation. Bul. Ent. Soc. Amer. 14(3):210. (SP-30)
- Jay, Edward, Davis, Robert, and Brown, Sharon. 1968. Studies on the predacious habits of Xylocoris flavipes (Reuter) (Hemiptera: Anthocoridae). Jour. Ga. Ent. Soc. 3(3):126-130. (Sp-General)
- Laudani, H., and Tilton, E. W. 1968. USDA research program and facilities for the use of gamma irradiation in the control of stored-product insects.

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- Smithwick, Eleanor B. and Brady, U. Eugene. 1968. Gland ultrastructure and relative sex attractant concentrations in selected ages of female Indian-meal moths. Bul. Ent. Soc. Amer. 14(3):205. (SP-31)
- Soderstrom, Edwin L. 1968. Phototactic response of adults of confused flour beetle and red flour beetle and larvae of black carpet beetle to flashes of blue-white light. Jour. Econ. Ent. 61(4):973-975. (SP-57)
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- Wojcik, Daniel P. 1968. Tests for audible and ultrasonic sound production by stored-product insects. Jour. Econ. Ent. 61(5):1414-1417. (SP-57)

# Improved Insecticidal Control

- Anonymous. 1969. Fumigating crated cereal products. Agricultural Research (USDA) 17(8):16. (SP-61)
- McDonald, L. L. 1968. Relative effectiveness of Tropital and piperonyl butoxide as synergists for pyrethrins against stored-product insects. Jour. Econ. Ent. 61(6):1645-1646. (SP-61)
- Speirs, Roy D., and Zettler, J. Larry. 1969. Toxicity of three organophosphorus compounds and pyrethrins to malathion-resistant <u>Tribolium</u> castaneum (Herbst) (Coleoptera, Tenebrionidae). Jour. Stored Prod. Res. 4(4):279-283. (SP-61)
- Stern, William L., Gillenwater, Hagan B., Eason, Gerald, Garcia-Quintana, A., and Cail, R. S. 1968. Lindane and dichlorvos for protection of herbarium specimens against insects. Taxon 17 (6):629-632. (SP-61)

# Insect-Resistant Packaging

Anonymous. 1969. USDA experimental bags tested in overseas shipment. The Northwestern Miller 276(1):51-52. (SP-60)

- Highland, Henry A., Van Byrd, Richard, and Secreast, Margaret. 1968. Effect of ethylene vinyl acetate on the migration of piperonyl butoxide from coatings of synergized pyrethrins on kraft paper. USDA-ARS 51-28. (SP-59)
- Highland, H. A., Guy R. H., and Laudani, H. 1968. Polypropylene vs. insect infestation. Modern Packaging 41(12):113-115. (SP-59)
- Highland, Henry A., Secreast, Margaret, and Merritt, P. H. 1968. Polyvinylidene-coated kraft paper as an insecticide barrier in insect-resistant packages for food. Jour. Econ. Ent. 61(5): 1459-1460. (SP-59)

# Simplified Residue Analysis

- USDA, ARS Information. 1968. Portable gas chromatography. Agr. Res. 17(6):14. (SP-9(C8289))
- Haas, Gerhard J., and Guardia, Enrique J. 1968. Production of antibodies against insecticide-protein conjugates. Proc. Soc. Expt. Biol. Med. 129(2):546-551. (SP-11(C8880))

# General

- USDA, ARS. 1968. Addendum No. 1, Agriculture Handbook No. 331. Suggested guide for the use of insecticides to control insects affecting crops, livestock, households, stored products, forests, and forest products. (SP-General)
- National Academy of Sciences. 1969. Contributions to Chapter X, Cultural Control of "Principles of plant and animal pest control--Vol. 3, Insect-pest management and control." Publication 1695. (SP-General)

# PROGRESS REPORT OF THE INSTRUMENTATION RESEARCH LABORATORY MARKET QUALITY RESEARCH DIVISION AGRICULTURAL RESEARCH SERVICE

#### AREA 14

#### INSTRUMENTATION FOR OBJECTIVE MEASUREMENT

# USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1969
Location of Intramural Work	: Research Problem Area :
	: 501 : Total
Maryland (Beltsville)	: : 5.0 : 5.0
Total	: : 5.0 : 5.0

Intramural program is supplemented by extramural support representing (a) 4.0 SMYs at State Agricultural Experiment Stations and (b) 1.0 SMY at other U.S. institutions.

# Progress - USDA and Cooperative Program

### RPA 501 - IMPROVEMENT OF GRADES AND STANDARDS

1. <u>Electrical Properties</u>. Equipment has been assembled and techniques have been standardized for measuring electrical properties of apples over the frequency range from 100 K Hz to 1000 M Hz. (IL-3(G))

# 2. Light Transmittance Techniques.

- a. <u>Wide Range Spectrophotometer</u>. A wide range, high precision spectrophotometer has been constructed to provide true absorbance curves of dense light scattering samples over the wavelength range from 300 to 2300 nm. Measurements with this unit were used to develop a method for rapid analysis of oil, moisture, protein and starch content of soybean flour. (IL-6)
- b. <u>Pigment in Flower Petals</u>. A low cost microspectrophotometer has been constructed for absorbance studies of individual plant cells. Measurements on epidermal cells show anthocyanins in the cell to exist in a state different from that predicted by previous studies of

extracted pigments. The effects of pH, pigment concentration, and co-pigments on the absorbance properties of anthocyanins have been characterized. These studies help to explain the color and color changes of plant products. (IL-6)

- c. <u>High Energy Monochromator</u>. Design has been completed for a large area grating monochromator of the Czerny Turner type. The design goal is for a device providing at least 10 mw of radiant power at 700 nm with a bandpass of 10 nm.
- d. <u>ATR Technique</u>. The major cause of errors in ATR measurements on powdered samples has been identified as surface contact problems between the sample and the ATR crystal. A new sample holder has been constructed to permit a study of the effect of pressure and particle size on the surface contact. (IL-5(A))
- 3. Automatic Fruit Sorter. The conceptual design for an automatic machine has been completed. A prototype device for orienting apples has been constructed and tested. This device requires an average of 30 seconds to give satisfactory orientation to an apple. A prototype photometer has been constructed but tests show poor performance. The changes needed in the photometer design have been determined. (IL-7(C))
- 4. <u>Sonic Technique</u>. Sonic resonance tests for apple firmness correlated well with objective and sensory firmness tests during the harvest season. During 6 months storage, sonic measurements indicated that the late harvested fruit increased in firmness whereas the firmness of early harvested fruit decreased. Correlations between sonic tests and panel scores for firmness of apples after storage were poor. (IL-6)

## Publications - USDA and Cooperative Program

Light Transmittance Techniques

- Asen, S., K. H. Norris, and R. N. Stewart. 1969. Absorption spectra and color of aluminium cyanidin 3-glucoside complexes as influenced by pH. Phytochemistry 8:653-659. (IL-6)
- Ben-Gera, I., and K. H. Norris. 1968. Determination of moisture content of soybeans by direct spectrophotometry. Israel J. of Agr. Research 18(3):125-132. (IL-6)
- Ben-Gera, I., and K. H. Norris. 1968. Influence of fat concentration on the absorption spectrum of milk in the near infrared region.

  Israel J. of Agr. Research 18(3):117-124. (IL-6)

- Beroza, M., K. R. Hill, and K. H. Norris. 1968. Determination of reflectance of pesticide spots on chromatographs using fiber optics. Analyt. Chem. 40(17):1608-1613.
- Bittner, D. R., and K. H. Norris. 1968. Optical properties of selected fruits versus maturity. Transactions of American Society of Agr. Engineers 11(4):534-536. (IL-6)
- Norris, K. H. 1968. Evaluation of visible radiation for plant growth.

  Annual Review of Plant Physiology 19:490-499. (IL-6)
- Stewart, R. N., S. Asen, K. H. Norris, and D. R. Massie. 1969.
  Relation of flower color to optical density spectra of intact tissue and of anthocyanin extracts. American J. of Botany 56(2):227-231.

  (IL-6)

## Sonic Technique

Abbott, J. A., N. F. Childers, G. S. Bachman, J. V. Fitzgerald, and F. J. Matusik. 1968. Acoustic vibration for detecting textural quality of apples. Proc. American Society for Hort. Sci. 93:725-737. (IL-6(C))

# PROGRESS REPORT OF THE

PIONEERING RESEARCH LABORATORY
MARKET QUALITY RESEARCH DIVISION
AGRICULTURAL RESEARCH SERVICE

#### AREA 15

#### PIONEERING RESEARCH

## USDA and Cooperative Program

Location of Intramural Work	:	Scientist Man Years, FY 1969
Maryland (Beltsville)	:	2.2
Total	:	2.2

## Progress - USDA and Cooperative Program

RPA 404 - QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES

## A. Postharvest Physiology

- 1. Ethylene formation. The ethylene forming enzyme, methional peroxidase requires prior reaction with codecarboxylase and also Mn<sup>2+</sup> as cofactor. A two-step reaction which converts the amino acid to an aldehyde or similar intermediate, as a first step and a methional-cleavage reaction as a second step is dependent on the two reaction systems being separated within the cell. The first system may be mediated by an Mn<sup>2+</sup> dependent codecarboxylase and the second system requires hydrogen peroxide and a free radical to cleave methional or a closely related intermediate. Evidence is being accumulated for the postulation. (PL-1)
- 2. Ethylene and enzyme formation. Reports that ethylene induces formation of enzymes such as peroxidase, polyphenolase, and phenylalanine ammonia-lyase have led to studies with pea seedlings and apple tissues, which indicate that ethylene causes changes in amounts of peroxidase isoenzymes and also some change in the isoenzyme patterns. Techniques are still being developed to follow this promising method for determining subtle changes in enzyme patterns after treatment with ethylene. (PL-1)
- 3. Cyclic adenosine monophosphate-phosphodiesterase in plant tissue. The recent discovery by others that 3',5' cyclic adenosine monophosphate (AMP) is the mediator of numerous hormone effects in animal

systems has led us to succeed in demonstrating the presence of 3',5' cyclic AMP-phosphodiesterase in tissues of seedlings and fruits. We have not yet been successful in unequivocally demonstrating the presence of cyclic AMP in plant tissues. If this system is present in plants and operates as a mediator of the action of plant hormones its study should have great significance in elucidating many problems in plant metabolism. (PL-1)

4. <u>Ultrastructure of fungus spores</u>. Thermal death of germinating sporangiospores of <u>Rhizopus stolonifer</u>, and conidiospores of <u>Monilinia fructicola</u>, was not linked to a single first event at the ultrastructural level. Three effects always observed after 2.5 minutes at 125° F. were: (1) formation of ribosomal aggregates; (2) disruption or derangement of mitochondria; (3) disruption or derangement of nuclei.

Others have determined that during germination of the <u>Rhizopus</u> sporangiospore, a thin inner wall is formed and this eventually stretches to form the wall of the germ tube as it emerges from the spore. We have observed that during inner-wall formation groups of globular-shaped bodies containing a crystalline-like arrangement of particles about 50-60 Å diameter were present and are perhaps involved in the synthesis of the inner wall.

The <u>Monilinia</u> conidiospore wall is composed of two layers, a thin layer and a thick, electron transparent inner layer. There were numerous organelles resembling microbodies, and globular bodies, presumably lipid storage bodies, present in the cytoplasm. (Cooperative with HCRB) (PL-2)

- 5. Purification of cytochrome b-559 from mitochondria of mung bean seedlings. Cytochrome b-559 purified earlier from sweetpotato mitochondria has also been obtained from mung bean mitochondria. These results are very important in clarifying the complex picture of b-type cytochromes in plant mitochondria and help establish that mung bean mitochondria have the same complement of b-cytochromes as do other plant mitochondria and that the b-559 component occurs generally in plant mitochondria. (PL-2)
- 6. <u>Ultrastructure of barley and wheat embryos</u>. Mitochondria begin to show loss in structural integrity as wheat and barley seeds dry on the plant. Many other organelles remain intact. Mitochondria in the embryo of the dry seed show little internal organization, although the outer membrane appears intact. Studies are under way to determine at what time during germination intact mitochondria appear. (Cooperative with FCAP)

## Publications - USDA and Cooperative Program

- RPA 404 QUALITY MAINTENANCE IN MARKETING FRUITS AND VEGETABLES
- Baker, J. E., L. G. Elfvin, J. B. Biale and S. I. Honda. 1968. Studies on ultrastructure and purification of isolated plant mitochondria. Plant Physiology 43: 2001-2002. (PL-2)
- Fuchs, Y. and M. Lieberman. 1968. Effects of kinetin, IAA, and gibberellin on ethylene production and their interactions in growth of seedlings. Plant Physiology 43: 2029-2036. (PL-1)
- Hruschka, H. W., W. L. Smith, Jr., and J. E. Baker. 1969. Reducing chilling injury of potatoes by intermittent warming. Amer. Potato J. 46(2): 38-53. (PL-2)
- Lieberman, M. and A. T. Kunishi. 1968. Origins of ethylene in plants. Phytopathological Society of Japan, 165-179. (PL-1)
- Takeo, T. and M. Lieberman. 1968. 3-methylthiopropionaldehyde peroxidase from apples: An ethylene-forming enzyme. Biochimica et Biophysica Acta 178: 235-247. (PL-1)

# PROGRESS REPORT OF THE COLOR RESEARCH LABORATORY MARKET QUALITY RESEARCH DIVISION AGRICULTURAL RESEARCH SERVICE

#### AREA 16

#### COLOR RESEARCH

## USDA and Cooperative Program

	: Scientist Man-Years F.Y. 1969
Location of Intramural Work	: Research Problem Area : 501 : Total
Maryland (Beltsville)	: : 1.0 : 1.0
Total	: : 1.0 : 1.0

No extramural support

#### Progress - USDA and Cooperative Program

1. <u>Colorimetry</u>. External color measurement and preference judgment of Wolcott blueberries with bloom removed was significantly different from fruit with bloom. Mean rank scores of 28 anelists showed bloom fruit as most preferred. Anthocyanin extractions of individual fruit correlated highly with light transmission measurement of intact berries whether  $\frac{4}{2}$  OD (710-800nm) or  $\frac{4}{2}$  OD (740-800 nm) was used.

Differences in spectral data and colorimeter measurement of normal and faded cranberry beans served to initiate a cooperative study with the Grain Division, Consumer and Marketing Service, on accelerated color fading using xenon light. Accelerated fading with xenon is evaluated concurrent with color change under continuous and intermittent fluorescent light. Preventive measures to reduce color fading in handling grain standards is the objective.

2. Appearance Evaluation. Potato chips viewed under 7500K fluorescent fixtures having reflectors which both included and excluded UV energy did not appear different to panelists although instrument measurement was affected by the fluorescence of the chips. Color measurement of flat chips showed significant increase in the Hunter  $\mathbf{b_L}$  values when UV light was included in the measuring beam. At the same time,  $\mathbf{L}$  and

- a<sub>L</sub> values decreased slightly. A new photographic color guide of both flat and curly chips is being made by Munsell Color Company; color data and visual ratings of all chips in the guide are known.
- 3. <u>Spectroradimetry</u>. Spectroradiometer measurement of light fixtures showed spectral curve differences due to operator usage, light collector (sphere or cosine receptor) and/or phototube aging. Lights visually unlike but both ca. 7500K were spectrally unlike at 630 and 660 nm.
- 4. <u>Computer Systems</u>. Spectral data handling system for IBM 360, Model 30 was completed. Data flows through NCR punched-magnetic tape converter, to X, Y, Z conversion (Rensselaer program) then to Munsell conversion (NBS program). Where necessary data may flow alternative routes for light scattering analysis (CRL-DSAD program) and calculation of MacAdam color difference (Rensselaer program). Complete data handling may be done in the Color Research Laboratory on programmable calculators with the exception of the Munsell conversion. (CL-1)

# Publications - USDA and Cooperative Program

Yeatman, J. N. 1969. Tomato products: read tomato red? Food Tech. 23(5):20-29. (CL-1)

